How many vPvM/PMT substances have been registered under REACH? - vPvM/PMT screening by using the Danish QSAR database

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SETAC Europe 28th Annual Meeting
13–17 May 2018 | Rome, Italy

ABSTRACT BOOK

Responsible and Innovative Research for Environmental Quality
ABSTRACT BOOK
SETAC Europe 28th Annual Meeting

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This book compiles the abstracts from the platform and poster session presentations at the 28th Annual Meeting of the Society of Environmental Toxicology and Chemistry - Europe (SETAC Europe), conducted at the Rome Convention Centre La Nuvola, Rome, Italy, from 13 – 17 May 2018.
The abstracts are reproduced as submitted by the author and accepted by the Scientific Committee. They appear in order of abstract code and alphabetical order per presentation type. The poster spotlight abstracts are included in the list of poster abstracts. The presenting author of each abstract is underlined.

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SOCIETY OF ENVIRONMENTAL TOXICOLOGY AND CHEMISTRY
In the 1970s, no forum existed for interdisciplinary communication among environmental scientists, biologists, chemists, toxicologists, managers, engineers or others interested in environmental issues. The Society of Environmental Toxicology and Chemistry (SETAC) was founded in North America in 1979 to fill the void, and quickly saw dynamic growth in the Society's membership, meeting attendance and publications.

A unique strength of SETAC is its commitment to balance the scientific interests of government, academia and business. The Society by-laws mandate equal representation from these three sectors for officers of the World Council and Geographic Unit Boards of Directors and Councils, and in the composition of committees and other society activities. The proportion of members from each of the three sectors has remained nearly equal over the years.

The Society is concerned about global environmental issues. Its members are committed to Environmental Quality Through Science®, to timely and effective communication of
research, and to interactions among professionals so that enhanced knowledge and increased personal exchanges occur. Therefore, SETAC publishes two globally esteemed scientific journals and convenes annual meetings around the world, showcasing cutting-edge science in poster and platform presentations. Because of its multidisciplinary approach, the scope of the science of SETAC is broader in concept and application than that of many other societies.

SETAC’s growth is reflected in the founding of geographic units around the world. SETAC Europe was established in 1989 as an independent organisation, followed by SETAC Asia-Pacific in 1997 and SETAC Latin America in 1999. In 2002, the four existing organisations joined together under the governance of the SETAC World Council. SETAC Africa is the most recent geographic unit, which was adopted in 2012. As evidence of international acceptance of the SETAC model and of the great interest at the local level, regional chapters and branches have emerged in a number of countries.

SETAC publishes two journals: *Environmental Toxicology and Chemistry* (ET&C) and *Integrated Environmental Assessment and Management* (IEAM). Environmental Toxicology and Chemistry is dedicated to furthering scientific knowledge and disseminating information on environmental toxicology and chemistry, including the application of these sciences to risk assessment. Integrated Environmental Assessment and Management focuses on the application of science in environmental decision-making, regulation, and management, including aspects of policy and law, and the development of scientifically sound approaches to environmental problem solving. Together, these journals provide a forum for professionals in academia, business, government, and other segments of society involved in the use, protection, and management of the environment for the enhancement of ecological health and human welfare.

SETAC books provide timely in-depth reviews and critical appraisals on scientific subjects relevant to understanding a wide range of contemporary topics pertaining to the environment. These include any aspect of environmental chemistry, toxicology, risk assessment, risk management, or environmental policy.

SETAC has two administrative offices, in Pensacola, Florida, USA, established in 1992, and in Brussels, Belgium, established in 1993.
Keynote abstracts

Keynote Sunday
Responsible Research and Innovation (RRI) - a Path towards Sustainability?
Elizabeth Stranack, University of Bergen, Centre for the Study of the Sciences and the Humanities, Norway

Responsible Research and Innovation (RRI) is a cross-cutting principle of EU’s research funding programme “Horizon 2020”. Indeed, in Rome 2014, scientists and policy-makers jointly produced the “Rome Declaration on RRI in Europe”, that states that “excellence today is about more than ground-breaking discoveries – it includes openness, responsibility and the co-production of knowledge”. The principle of RRI acknowledges that civil society is entitled to “speak back” to science and help shape the knowledge and technology of tomorrow in an ethically acceptable and sustainable direction.

What does RRI entail in practice, for researchers, innovators and policy-makers? How could RRI principles, indicators and practices help to pick up more early warnings to avoid costly late lessons from unfortunate impacts of science and technology? The lecture will present the conceptual basis of EU’s RRI policy. Specifically, a full appreciation of RRI depends on a theoretical understanding not only of risk, but also of decision-making under uncertainty, ignorance and indeterminacy.

Keynote Monday
Food Safety in a Complex Changing World
Bernhard Utl, EFSA, Italy

EFSA provides independent scientific advice on all matters related with food and feed with a direct or indirect impact on human, plant and animal health. Effects on the environment are also considered as they may pose an indirect risk to food and feed. EFSA takes into account environmental risk assessment in its assessments of the application of plant protection products, the deliberate release into the environment of GMOs and the use of certain substances in food and feed (e.g. feed additives). EFSA also assesses the environmental risks related to the entry and spread of invasive alien species harmful to plant health.

EFSA is looking into the future, keeping up with a rapidly evolving and globalised world, characterised by dramatic environmental and other global changes (e.g. economic, political social, and technological) and an exponential growth and availability of data. These set new opportunities and challenges to the assessment of risks to both the environment and food safety and can drive their reemergence. In this context, EFSA is reviewing the methodologies for risk assessment and for the identification of emerging risks within its remit.

Predictive modelling tools based on holistic approaches for environmental risk assessment in realistic landscapes and under different scenarios of multiple stressors are being developed. Approaches considering the complex interactions and dynamics between the different food system actors, their behaviour and external drivers are proposed as tools useful for long term anticipation of emerging risks. Expert knowledge elicitation, horizon scanning, and crowdsourcing are being explored as tools to broaden participation, strengthen engagement of all relevant stakeholders and manage interconnectivity, in application of principles of resilience thinking.

Environmental quality and food safety are strongly intertwined. They need to be considered together when aiming toward the achievement of sustainable development goals. Consistent approaches for scientific assessment and data management need to be developed, integrating also societal, technological and economic drivers to effectively cope with the dramatic global changes and the data revolution we are observing.

Keynote Tuesday
Innovative Research Issues in Environmental Mutagenesis
Eugenia Dogliotti, Istituto Superiore di Sanità, Department of Environment and Health, Italy

During the 1920s, mutation research was put on a firm basis by H. J. Muller, who developed the concept of “mutation rate” and devised quantitative techniques for its measurement. These techniques allowed the discovery of the mutagenic action of ionizing radiation and paved the way for the pioneering work of C. Auerbach on chemical testing, starting with mustard gas. Since that time the recognition of the multitude of possible sources of mutagenic insults promoted the development of the science of environmental mutagenesis. Today the mechanisms by which chemicals induce mutation and the role of genetic susceptibility in the response to environmental mutagens have been largely explored. Moreover, a battery of test methods is available for regulatory purposes. What are the current challenges in environmental mutagenesis? New techniques for mutation research have been developed. The “omics” technologies such as whole genome sequencing, epigenetic profiling, transcriptomics, proteomics and metabolomics have provided a snapshot of the effects of genetic polymorphisms, gene regulation, protein synthesis and stability, metabolic pathways in the control of cell function. This presentation will describe: 1) the successful identification of the mutagenic environmental agents underlying certain types of cancer by using whole genome sequencing; (ii) the evidence that epigenetic alterations mediate toxicity from environmental chemicals and, (iii) the use of the exposome approach, that comprises all environmental exposures that a person experiences from conception throughout the life course, to unravel complex gene environment interactions that affect disease risk.

Keynote Wednesday
The Environmental Dimension of Antimicrobial Resistance: Assessing and Managing the Risks of Anti-infectives
Jason Snape, AstraZeneca Global Safety, Health and Environment, UK

Antibiotics are vital in the treatment of infectious disease in both livestock and human health and they are entering the environment continuously. In freshwaters antibiotics can reach concentrations up to mg/L, but more commonly they occur in the low to sub μg/L range. They selectively target bacteria and thus there is an increased likelihood for impacts on environmental bacteria populations at levels well below that for effects on aquatic vertebrates. However, current environmental risk assessment (ERA) frameworks of antibiotics, as required by the European Medicines Agency guidelines 2006, adopts the use of one species of cyanobacteria only to represent all bacterial diversity. The activated sludge respiration inhibition test (ASRIT), used to identify risk to microorganisms in sewage treatment plants has also been proven to be insensitive for antibiotics. Thus, there is concern that the ERA for antibiotics does not fully consider their potential impacts on microbial community structure, function and resilience. In addition to the risk posed to ecosystem function there is a global concern on antimicrobial resistance (AMR) development and the associated risk to human health. It has been proposed that the risk of AMR development in the natural environment should be included in ERA but there is currently no standard experimental methodology or framework to address this. Recently, a theoretical approach that makes use of minimum inhibitory concentrations (MIC) of clinically relevant bacteria (CRB; using the European Committee on Antimicrobial Susceptibility Testing (EUCAST) database) has been proposed to predict no effect concentrations (PNEC) for AMR development (PNECR). To help define science-based protection goals for antibiotics for use in a prospective ERA frameworks and to define safe discharge concentrations for antibiotic production and patient use this presentation will review the publicly available aquatic ecotoxicity data for antibiotics to assess the following: 1) the relative sensitivity of commonly used taxa in aquatic ecotoxicity to antibiotics; 2) the value of extending the toxicity testing to a more diverse range of bacteria species and; 3) how a PNEC relates to the PNEC derived for surface waters (PNECSW) using standard ecotoxicity testing. This presentation will describe (i) the output of this analysis of protection goal data and (ii) how the wider pharmaceutical industry are addressing concerns with antibiotic residues associated with manufacturing operations.
Modelling and monitoring of pesticides fate and exposure in a regulatory context (I)

1 The SETAC DRAW workshops - aims, approaches and progress to date


In order to develop a more complete understanding of spray drift to improve the regulatory basis for representation in risk assessments, a set of SETAC workshops known as DRAW (Drift Risk Assessment Workshops) are underway to facilitate a range of efforts: Assemble and interpret a database of the spray drift trials for boom sprayers; Develop a programme of trials to more fully characterise drift influencing factors; Use this information to develop proposals for standardized protocols for drift characterization in the field Develop an enhance role for mathematical modelling as a higher tier risk assessment option; and Expand and reinforce the toolbox of regulatory risk mitigation measures. This presentation will focus upon; Database development and study design The workshop database currently comprises 56,001 data points from a wide range of studies, providing a rich, complex basis for supporting a range of different research efforts. Best practices for the variation in study designs and the consequent difficulties with interpretation there is a clear motivation to develop a more detailed and tightly defined protocol to support future research efforts. This presentation will summarise the database and efforts to develop and test a protocol to support further research efforts. Modelling Options for developing an expanded role for modelling of drift profiles have focussed upon evaluations of two models that have been used within a regulatory context in the EU; IDEFICS and the SSAU Arable Crop Spray Drift Model. This presentation will report on model assessment efforts, potential future improvements in process representation and consider options for regulatory scenario development. Flexibility in risk mitigation An earlier workshop (SETAC MAgPIE) compiled a toolbox of risk mitigation measures in use in Europe and recommended development of communication tools to support broader and more effective implementation and encourage certification and testing harmonisation. This presentation will summarise the efforts underway in SETAC DRAW to realise these objectives through the creation of a platform to support exchange on scientific, technical, professional, and legislative or regulatory aspects of the toolbox, to further develop its accuracy and effectiveness (https://www.spraydriftmitigation.info/).

2 Plant uptake in regulatory environmental exposure assessment: Refined modelling based on experimental data

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A novel study design to determine plant uptake of chemicals for environmental fate modelling was developed and tested in a tiered approach. Ten laboratory organizations each performed a series of experiments with uptake testing participated in a round robin test and studied uptake of [14C]-1,2,4-triazole by wheat plants. Afterwards, uptake of ten radioactively labelled chemicals with various properties by potato, tomato or wheat plants was investigated in two laboratories. The findings showed acceptable inter-laboratory variability and proved the applicability of the design to various compound/crop combinations. Experimental Transpiration Stream Concentration Factor (TSCF) values were higher than calculated values, implying that the equation of Briggs et al. (1982) underestimates plant uptake of compounds with logKow values of less than 2. Results obtained with this study type are suggested to be used for regulatory environmental exposure assessments. These schemes are usually based on a tiered approach, where modelling with refined model parameters is one of the higher tier options. One example is the plant uptake factor that is considered e.g. in the course of the leaching assessment according to FOCUS Groundwater. Consideration of plant uptake (dissolved compound mass is taken up into plants with the soil porewater) decreases compound mass in soil and can lead to more realistic predicted environmental concentrations. Recent guidance proposes a default uptake factor of 0 for modelling and suggests two refinement options. For TSCF values within a certain range, use the value derived from the equation of Briggs et al. (1982) and the substance specific TSCF value from “uptake experiments with appropriate and agreed set-up to be developed” (EFSAs, 2013; EU Com, 2014). The presented study design was explicitly developed to address the need identified by EFSAs and was based on suggestions from the EuregPuf workshop (York, 2015) where participants from academia, authorities and industry met to establish an up to date understanding of plant uptake science. Lessons learned from the testing and continuous exchange with academia and authorities facilitated optimisation of the study design. The current version of the study design is considered appropriate to produce reliable data on plant uptake to be used as input for refined exposure modelling. An explicit guidance, however, on how to integrate the requested study design into the regulatory process is still lacking.

3 Work of a SETAC Group to Develop the Scientific Basis for Guidance for Regulatory Groundwater Monitoring of Crop Protection Products and their Metabolites in Europe

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Groundwater monitoring is considered a higher tier assessment in the regulatory groundwater assessment of crop protection products in Europe, but little guidance is available to date in this study design. The SETAC EMAG-Pest GW group (a mixture of regulatory, academic, and industry scientists) in 2015 began developing the scientific basis for guidance for use by regulators and industry scientists. Rigid study designs are not appropriate since the study design needs to be tailored to the specific study objectives and should consider environmental conditions, the properties affecting environmental behaviour of the substance being studied, and site and use conditions. To illustrate how study design can vary, the group has proposed general study designs for seven hypothetical exposure assessment options, ranging from protecting all zones of saturation below the soil surface to only groundwater used to supply drinking water. Designs include recommendations on in-field and edge of field studies, as well as studies focused on catchments and aquifers. Examples of potential study designs and recommendations on the use of publicly available monitoring data have also been included. Also general recommendations on well installation and sampling procedures have been provided. Methodology has also been developed for assessing the relative vulnerability of agricultural regions and the vulnerability of specific sites for use in study design and site selection. The work of SETAC EMAG-Pest GW is still in progress but the goal is to finish by mid-2018.

4 Effect of the Freundlich exponent on the finite penetration depth in a homogeneous Freundlich-SFO leaching system

J. Boesten, Wageningen Environmental Research

All models used in the EU pesticide leaching assessment since 2000 (PELMO, PEARL, PRZM and MACRO) are based on a Freundlich isotherm combined with a single first-order (SFO) degradation of the pesticide concentration in total soil. This is one of the cornerstones of the EU regulatory leaching assessment. This assessment is based on the FOCUS groundwater scenarios which use weather series of tens of years and include crop development and heterogeneous soil profiles. The sensitivity of the FOCUS leaching concentration (evaluated at 1 m depth) to the parameter describing the curvature of the Freundlich isotherm (i.e. the Freundlich exponent N) as derived from simulations with these models shows a sharp decline with decreasing Freundlich exponent with the concentration going down to a submolecular level. This is counterintuitive and difficult to understand. Explanations may be found by studying a simplified version of these sophisticated models, i.e. a assuming a homogeneous soil profile with pesticide properties that are constant with depth and assuming a constant water flow rate and a constant volume fraction of water (further called ‘simplified Freundlich-SFO system’). Previously it was shown that a pulse of pesticide applied at the soil surface in this simplified system has a finite leaching depth beyond which no pesticide molecule ever pass. Simulation of experimental data for a few FOCUS groundwater scenarios were compared to this effect on the percentage leached with this simplified model and qualitatively these effects were found to be similar. Next it was shown that this finite penetration depth after infinite time in the simplified Freundlich-SFO system increases slowly when N increases from 0.5 to about 0.85; however, when N approaches 1, this finite penetration depth goes to infinity. This was expected because this finite penetration depth does only occur in a system with a Freundlich isotherm and not in a system with a linear isotherm. It was checked by inspection of a concentration profile of one of the FOCUS groundwater scenarios that these scenarios also show a finite penetration depth for low N values at the end.
5 Bespoke monitoring to support Tier 4 FOCUS groundwater assessment

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Data generated from this bespoke groundwater monitoring programme will offer a solution to address the non-relevance case of pinoxaden metabolites from an exposure side. Median modelled mass flux was determined using GeoPEARL 3.3.3 simulations over 20 years which represent vulnerability to leaching across the EU27 under standard conditions. These data were aggregated to a 10km² level and combined with a shallow groundwater dataset and a cereal land use dataset based on wheat in CAPRI. Those grid cells in the upper 50th percentile for each spatial layer (mass flux, shallow groundwater, and wheat) were considered for the site selection process. Sites identified by modelling were assessed during site walkover surveys. To justify inclusion in the programme, sites had to have a history of pinoxaden use, groundwater less than 10m bgl, no confining layers, and no influence features which may act as preferential flow pathways. In 2015, 70 sites were installed across France, Germany, Italy, Lithuania and the United Kingdom. Each site consisted of three shallow wells installed around the field perimeter. The 70 sites represent all EU FOCUS groundwater scenarios except Jokioinen. The sites have all had a minimum of two pinoxaden applications before 2016 with groundwater levels an average of 2.9m below ground level. Sampling began in 2015 from 84 down hydraulic gradient wells. Of the 871 samples collected between June 2015 and July 2017 from these 70 vulnerable sites, the average for each site never exceeded 0.1µg/L. Only minor residues of metabolites have been detected since sampling began. Monitoring is to continue until Q4 2019 to ensure a thorough assessment of groundwater vulnerability is made. The modelling approach should allow extrapolation of the modelled vulnerability to be extended to member states outside of those where the wells were installed.

6 Long-Term Trend of Aquatic Pesticide Risk

A. Paulus, UFZ - Helmholtz Centre for Environmental Research / System-Ecotoxicology; S. Kullmann, K. Foit, Helmholtz Centre for Environmental Research UFZ / System-Ecotoxicology; M. Klein, Fraunhofer Institute for Molecular Biology and Applied Ecology IME; M. Liess, UFZ Centre for Environmental Research / System-Ecotoxicology

European Union member states aim at reducing ecological risks exerted by pesticides. For this, reliable trend indicators of pesticide exposure and risk are inevitable. Based on this demand we designed a long-term trend indicator of aquatic pesticide risk for Germany. It uses pesticide sales statistics, toxicity data and chemical properties as input variables. The trend indicator was designed by combining the most reliable exposure and effect models. (i) We selected the most frequent exposure model by evaluating several established exposure models. For this we tested their performance with peak concentration data monitored in small agricultural streams in central Germany. These field data comprised of event-driven samples of 46 active substances from field-campaigns performed in 1998, 1999, 2000, and 2013. The highest agreement of measured and modelled peak concentration was yielded by the risk indicator EXPOSITIVE (R²: 0.38), followed by the more complex models POCUS STEP 2 (R²: 0.36), SYNOPS-TREND (R²: 0.24), and GERDA (R²: 0.24). (ii) The translation from toxic pressure to pesticide risk was implemented by applying the field based and validated exposure − response relationship SPEARcolour. Based on these information and models, we calculated the trend of toxic pressure and pesticide risk in Germany from 1996 to 2016 for the 500 substances authorized in this period. The method presented here requires only few input data, is based on validated models and can be adapted to regional conditions around the world.

Hydrophobic Chemicals and Mixtures: Reliable Investigations on their Environmental Fate and Effects (I)

7 The hydrophobicity delay: symptoms and solutions

A. Celsius, Queens University; D. Mackay, Trent University / Chemistry; D. Powell, DMER Ltd.; J. Parnis, Trent University / Cemistry

The objective of this presentation is to set out the conditions under which chemicals of high hydrophobicity experience significant delays in approaching equilibrium conditions. We suggest that this delay may be misinterpreted as being caused by a change in partitioning behaviour or mechanism resulting in development of non-linear regression models describing inter-media partitioning. In reality, the partitioning is fundamentally linear but is distorted by a kinetic delay. The rate constant format commonly applied to fish biotaque from water of concentration Cº: Kow/Kw = Kow log Cº/Kw. This is a simple fish concentration, Kow is the uptake and loss rate constants and k is kBCF where BCF is the bioconcentration factor. The characteristic time for uptake and loss t is L^2/Kow/Kw. Slower uptake and loss will occur if the partition ratio Kow is large, and the fish must contact Kow-L its own volume to approach equilibrium. Very hydrophobic substances will experience long time delays when approaching equilibrium and correspondingly long times for loss during a deputation phase. Mackay et al. [1] modeled bioconcentration and toxicity of superhydrophobic chemicals D4, D5, and D6 using a biotakue model for fish. Due to the very high hydrophobicity (log Kow=10³) for D5 and very low water solubilities Cw must be very low, which results in a very long equilibration time. Uptake time to equilibrium for D5 was estimated to be ~2000 days, to get Cw=0.01mgl⁻¹ about 17 days. The study concluded that for superhydrophobic substances organisms will likely not reach toxic concentrations within the test duration which is usually 4-94h. Doutette et al. [3] reviewed foliage/air partitioning data in which the onset of a hydrophobic delay (HD) is approximately when log Kow=10³ develops - an HD problem for uptake of hydrophobic chemicals by foliage. This model shows levelling off corresponding to insufficient time to achieve equilibrium. A kinetically limited regime is reached at a Kow=10⁶ at the moment we will address the HD issue that we believe is a widespread phenomenon applicable to numerous environmental systems including passive sampling and partitioning to aerosol particles. Finally, we suggest a general method for identifying the HD problem. [1] Environ Sci Technol 2015, 49(19): 11913-22. [2] Environ Toxicol Chem 2012, 31(8):1911-9. [3] Environ Toxicol Chem. 2017, Accepted. [4] Environ Sci Technol 1999 33:1799-1804.

8 Partitioning of chlorinated paraffins (CPs) to organic matter is not class specific: implications for bioaccumulation

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Chlorinated paraffins (CPs) belong to a group of industrial chemicals consisting of n-alkanes (from 10 to 30 carbon chain atoms) with chlorine content from 30 to 70% weight. They are widely used as high-pressure lubricants, flame retardants, and additives in plastic, rubber, and sealants, leading to high-volume production worldwide. These chemicals are also ubiquitously found in the environment. The use of short chain chlorinated paraffins (SCCPs) in Europe has been restricted, however, medium (MCCPs) and long chain (LCCPs) chlorinated paraffins are used in Europe as substitutes for SCCPs. In some countries, all classes are still in use, leading to high production volumes (over a million tons per year globally). There is a lack of data on CP physicochemical and hazard-based properties, which is due to their inherent high complex. CPs are hydrophobic contaminants, which complicates their aquatic toxicity testing. In this work, we validate the use of passive dosing for the study of chlorinated paraffins and partitioning behavior of CP technical mixtures between silicone, water and organic carbon. We used 5 different technical mixtures from the three established categories (2 SCCPs, 1 MCCP, 1 LCCP). We added Daphnia magna to the passive dosing system, to understand the partitioning behavior of CP technical mixtures from CP-dosed water medium to CP-free organic matter (KOWwater). Bioaccumulation of D. magna was observed after 48 hours under different exposure concentrations. APCLI-QTOF-MS was used for CP quantification. Both silicone-water and organic carbon-water-partition coefficient overlapped between different categories of CP technical mixtures. CP-52, labelled as a MCCCP, had a similar silicone-water-partitioning coefficient as a restricted SCCP – Huel’s 70C. We demonstrate that increasing average chlorine content of each CP mixture significantly increases the Log KOWwater and Log KOWwater. These results could have implications on the study of environmental fate of CPs: in-use CPs (MCCPs and LCCPs) might be equally or more bioaccumulative as restricted SCCPs. KOWwater is particularly helpful at predicting bioaccumulation of chemicals into biota. The next step is to quantify the bioaccumulation potential of CPs. With the use of the passive dosing approach, we are producing laboratory experimental data that can be used to help in the on-going regulatory discussion on MCCPs and aid their risk assessment.

9 Trophic magnification of cyclic volatile siloxane materials (D4, D5, and D6) in a freshwater lake: A Monte-Carlo analysis


The trophic transfer of cyclic methylsiloxane (cVMS) materials in aquatic ecosystems is an important criterion for assessing bioaccumulation and ecological risk. These compounds are known to be bioaccumulative, specifically octamethylcyclotetrasiloxane (D4), decamethylcyclopentasiloxane (D5), and dodecamethylcyclohexasiloxane (D6) was determined for the Lake Pepin, Minnesota (USA) food web. The objective of this work was to determine if cVMS materials are biomagnified in this freshwater ecosystem. To determine whether the benthic influence in the Lake Pepin aquatic food web affected the trophic magnification factor (TMF) values for the cVMS compounds, a companion study was conducted to determine the biomagnification and TMF value of a reference material, 2,3,3',4,4',5,5'-heptachlorobiphenyl (PCB-180), in Lake Pepin. TMFs for the three cVMS materials and PCB-180 were determined using standard
methods involving feeding guild, trophic guild classifications, and the stable isotopes of nitrogen (δ15N) and carbon (δ13C) to estimate trophic position/carbon flow. The aquatic food web consisting of two benthic macroinvertebrate species and 15 fish species was evaluated for trophic magnification of cVMS materials and PCB-180. Lipid-normalized concentrations of D4, D5, and D6 were greatest in the lowest trophic levels and significantly decreased going up the food web, with the lowest concentrations occurring at the higher trophic levels. The TMFs measured for the three cVMS materials were almost all 99% for the uncertainty for cVMS TMF values in Lake Pepin was explained by uncertainty at the base of the food web (89%) and at the top of the food web (11%). BY comparison, PCB-180 had a TMF of 2.2 in the evaluated food web, indicating biomagnification. TMFs for the cVMS chemicals and PCB-180 were determined using a Monte-Carlo probability analysis technique, and the likelihood that the values exceeded unity was less than 0.5% for all three cVMS compounds and >99.5% for PCB-180. This evaluation indicates that D4, D5, and D6 do not biomagnify in the benthic-dominated Lake Pepin aquatic ecosystem, a food web which does demonstrate biomagnification of the legacy contaminant, PCB-180.

10 Distribution and Bioaccumulation of Polycyclic Aromatic Hydrocarbons in Aquatic Systems from the United States and China

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The present study reports the discovery of a suite of polycyclic aromatic hydrocarbons (PAHs) in aquatic sediments collected from four watersheds located in the United States and China, including the Gulf of Mexico (USA), San Francisco Bay (USA), Lake Tai (China), and Lake Dianhuan (China), and their bioaccumulation in the San Francisco Bay ecosystem. A total of 11 halogenated carbazoles, including 3-chloro, 3,6-dichloro, 1,3,6,8-tetrachloro, 2,3,6,7-tetrachloro, 3-bromo, 2,7-dibromo-3,6-dibromo, 3,6-dibromo, 1,3,6,8-tetrabromo-1-bromo-3,6-dichloro, and 1,8-dibromo-3,6-dichloro-carbazoles were screened. Halogenated carbazones were detected in 98.7% of the sediment samples, with concentrations ranging from below method limits of quantification to 51.5 ng/g dry weight. In most of these sediment samples, PHCZ concentrations exceeded those of polybrominated flame retardant (PBFRs). The latter group of chemicals has been demonstrated to be persistent and globally distributed. PHCZs were also detected in various organisms from the San Francisco Bay, including bivalves, sport fish, harbor seal blubber and bird eggs. The median concentrations of PHCZs by species ranged from 33.7 to 164 ng/g lipid weight. Biomagnification was also observed from fish to harbor seal and was mainly driven by chlorinated carbazoles, particularly 36-CCZ. Congener compositions differed among species, suggesting that individual congeners may be subject to different bioaccumulation or metabolism in species occupying various trophic levels in the studies system. Toxic equivalent (TEQ) values of PHCZs were determined on the basis of their relative effect potencies (REP) compared to 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD). The median TEQ ranged from 4.8 to 19.5 pg TEQ/g lipid weight in biological tissues. Our data demonstrated the broad exposure of PHCZs in the studies systems and potentially in global aquatic systems. These findings raise the need of additional research to better elucidate their sources, environmental behavior, and fate in global environments.

11 Bioconcentration factors of concentrates of essential oils in fish determined in an in vivo benchmarked dietary exposure study: A case study for pine oil

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Essential oils are fragrance materials that are registered as natural complex substances (NCS) under the European REACH legislation. One of the categories of information required in a REACH registration is information about the potential for bioaccumulation of NCS by fish. Determining the bioconcentration factor (BCF) of essential oils can result in reducing the standard flow-through uptake/deprivation experiment. Previously, we demonstrated that a single dietary exposure coupled to the benchmarking technique could be applied to an artificial mixture for measuring the in vivo BCF. Here, we report an application of our proposed BCF-determination methodology on a real essential oil – pine oil. Fish (rainbow trout) were dosed with a mixture of the pine oil and a suite of benchmark compounds via a single daily exposure. The median BCF (k\text{in}) in the fish soma (without GIT) for the key pine oil constituents are 0.134 d\text{1} (β-Caryophyllene, BCP) – 1.41 d\text{1} (BACE) and they were 0.0709 d\text{1} (HCB) – 0.517 d\text{2} (DCB) for the reference chemicals. The test compounds depurated faster from the soma than the GIT, making estimated whole-body depuration slower (conservative) compared to the soma only. HCB was the chemical most resistant to depuration via all three test compounds. Benchmarking to HCBC reduced the standard error of measured k\text{in,DM} from the soma for most of the chemicals, with k\text{in,DM} ranging from 0.001 d\text{3} (PCB52) to 2.98 d\text{1} (BACE). The apparent BCF (BCF\text{a}) values in soma for the key components in pine oil and the reference chemicals were in the range of 98.2 L kg\text{1} (BACE) – 1030 L kg\text{1} (BCP) – and 267 L kg\text{1} (DCB) – 1730 L kg\text{1} (HCB), respectively; while for the benchmarked BCF (BCF\text{a}) in soma, they are 46.3 L kg\text{1} (BACE) – 2570 L kg\text{1} (BCP), and 208 L kg\text{1} (DCB) – 197000 L kg\text{1} (PCB52) respectively. We conclude that a single dietary exposure coupled with the benchmarking technique is a feasible experimental approach for measuring the BCF of NCS in fish.

12 ROLE OF ADIPOSE TISSUE RESPONSIBLE FOR ECHOLOCATION IN THE BIOACCUMULATION PROCESS OF LIPOPHILIC COMPOUNDS IN HARBOUR PORPOISES

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Previous studies have suggested that persistent organic pollutants (POP) can lead to adverse effects in marine mammals, including harbour porpoises (Phocoena phocoena), thereby causing illnesses. Traditionally, blubber is an ideal matrix to assess POP bioaccumulation in marine mammals. However, during times of energy deficits, blubber tissue is broken down in which POPs are redistributed in the body. Echolocating tissues melon and mandibular fat are inert lipid bodies in odontocetes and, in contrast with blubber, are less prone to release POPs, which makes them ultimate sinks for POP lifetime bioaccumulation. This study aimed to assess the lifetime bioaccumulation of POPs in harbour porpoises through 1) analysis of POPs in various tissues and/or organs of harbour porpoises, including lipid rich bodies as blubber, menon and mandibular fat, and 2) Physiologically based toxico-kinetioc (PBTK) modelling of PCB 153 and PCBDE 153 to compare bioaccumulation of lipophilic compounds in lipid-rich tissues with different lipid composition and purpose (echolocation versus insulation) over the whole lifespan of male harbour porpoises. Overall, POP analysis and PCB 153 modelling for male harbour porpoises reveal that despite differences in lipid composition and lipid types, lipophilic pollutants bioaccumulation patterns are similar in blubber, mandibular fat and melon with increasing age. Nevertheless, the model showed the highest levels of POP 153 in mandibular fat, followed by melon and blubber. From these results, mandibular fat can be considered as a sink for PCB 153 and a better proxy for lifetime exposure than blubber, which can be both a sink and source of lipophilic pollutants. PCBDE 153 PBTK modelling reveals that bioaccumulation differs in lipid composition and lipid type, whereby bioaccumulation predominantly occurs in echolocating tissue during juvenile stage and in blubber during adulthood.

Keywords: Echolocation, life time bioaccumulation, biomonitoring, PBTK modelling, POP

Interpretation and uncertainty - overcoming challenges of translating LCA results into reliable information

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LCA: everything is relative and nothing is certain

LCA: everything is relative and nothing is certain

The first interpretation of 'correlation' h...
Drivers of variability and uncertainty in the chemical footprint of personal care products

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Chemical footprinting of products, quantifying the potential environmental impact of the product’s chemicals, could be used to inform consumers choice. However, the use of chemical footprints (ChFs) for comparative purposes requires a full understanding of the uncertainty and variability sources influencing its quantification. The goal of this work was to determine the ChFs for personal care products and quantify the variability and uncertainty in the different parameters used to derive these individual ChFs. In a first phase, we focused on shampoos. The environmental impact of each ingredient was derived from an environmental load, assuming 100% discharge to the drain, determined by the ingredient’s removal in activated sludge wastewater treatment plants (WWTPs) simulated using SimpleTreat, and a characterisation factor estimated with USEtox. The physico-chemical and ecotoxicological properties applied in both models were all estimated. Their reliability was derived from the prediction accuracy of the estimation models used (EPISuite, ACD Labs, ECOSAR). A Monte Carlo analysis with 1000 iterations was then performed, combining the uncertainty and variability of the different parameters, to determine the spread in ChFs. The ChFs derived by this approach spanned nearly 4 orders of magnitude (95% Confidence Interval (CI)). The wide span of the ChF’s 95% CI was primarily attributable to fragrances (61%), surfactants (20%), and the amount of products used (16%). The significant contribution from fragrances and surfactants can largely be explained by the uncertainty in their environmental impacts described by the characterisation factors derived with USEtox and more precisely the estimated ecotoxicity values. These preliminary results question the use of absolute values when communicating product’s chemical footprints. As long as more reliable ecotoxicological assessments are not available, identifying relative contributions to the overall environmental impacts might be more useful to target specific actions.

Combined uncertainty and scenario analysis within Life Cycle Assessment of waste management systems

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Life Cycle Assessment (LCA) is being increasingly used for decision support in the waste management field. LCAs are subject to uncertainty regarding both the input values (parametrical uncertainty) and its modelling choices (or epistemic uncertainty). Parametrical uncertainty can be systematically addressed with parametrical uncertainty analysis, while epistemic uncertainty can be addressed with scenario analysis. However, the communicability and usefulness of LCA’s results (16%) and the amount of products used (16%). The significant contribution from fragrances and surfactants can largely be explained by the uncertainty in their environmental impacts described by the characterisation factors derived with USEtox and more precisely the estimated ecotoxicity values. These preliminary results question the use of absolute values when communicating product’s chemical footprints. As long as more reliable ecotoxicological assessments are not available, identifying relative contributions to the overall environmental impacts might be more useful to target specific actions.

Which impact categories are relevant for LCA results interpretation?

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LCA is intrinsically a multicriteria approach comparing (almost) all the potential environmental impacts of human activities. However, multicriteria decisions pose challenges as a wide range of environmental impacts results may lead to unclear conclusions. Based on their relevance, a choice among the impact categories may be necessary. It can be carried out by examining how the information given in the Life Cycle Inventories (LCIs) is used by the impact categories. The Representativeness Index (RI) proposed by Esnouf et al. was initially used to compare the adequacy of Life Cycle Impact Assessment (LCIA) methods regarding LCIs. Here, the RI is used to explore the impact categories belonging to a given LCIA method. Thus, the present study focuses on how the interpretation of the LCA results can be undertaken given the choice of relevant impact categories. With a geometrical standpoint, LCIs of the ecoinvent database and impact categories of the ILCD method are standardized and localized within the same R² vector space. This vector space is generated by all the dimensions (i.e. elementary flows) from which the LCIs of the database are derived. The RI is a proximity measurement between any standardized LCI vectors and standardized impact category vectors, corresponding to the cosine of the angle between two vectors. This measurement does not assess the relevance of the environmental model behind impact categories, but rather translates the main elementary flows from an LCI based on how they are represented by the impact categories of an LCIA method. Two inventories referred to variability. Life Cycle Inventories (LCI) regionalization deals with increasing the geographic representativeness modelled in LCI. Life Cycle Impact Assessment (LCIA) regionalization deals with regionalized impact characterization that accounts for the spatial variability of the receiving environment. Regionalized characterization factors (CF) apply to spatialized elementary flows (EF), called LCI spatialization. However, integrating regionalization requires additional effort on data collection and treatment for LCA practitioners and database developers. Thus, prioritizing the regionalization effort on the most sensitive data (input data with uncertainty having the highest influence on the resulting uncertainty) would ensure an optimal use of resources to reduce LCA results uncertainty. This research work proposes a procedure to prioritize regionalization efforts based on global sensitivity analysis (GSA) to reduce the spatial uncertainty of LCA results. We applied this procedure to all the activities of two economic sectors (biofuel production and passenger land transport) defined in the ecoinvent database v3. The regionalized impact methodology IMPACT World+ is used to assess environmental impacts. Statistical tests are then used to derive sectoral recommendations regarding the impact categories (IC) and LCA phases (LCI or LCIA) that should be regionalized in priority. Those recommendations are meant to help LCA practitioners and LCI database developers to define their strategy for regional data collection to lower the LCA results uncertainty. Results show that contrasting IC ranking depending on the economic sector. For the biofuel production sector, land transformation encompasses almost all the uncertainty, whereas it is distributed among several impacts (global warming and marine acidification) on the land passenger transport sector. For LCA phases ranking, it confirms that inventory should be spatialized in priority for regionalized impact categories. This methodology allows providing different recommendations specific a sector to refine data collection in order to reduce uncertainty and enhance results interpretation. To our knowledge, this is the first time that an uncertainty analysis discriminating IC and LCA phase ranking is performed.

Poster spotlight: MO387, MO388, MO389

SETAC Europe 28th Annual Meeting Abstract Book
Environmental fate of emerging contaminants in the water cycle: analytical challenges and engineered solutions (I)

19
Unravelling longitudinal pollution patterns in freshwaters by non-target screening and cluster analysis
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Pollution of aquatic ecosystems with emerging organic contaminants (EOCs) has been intensively studied over the past decades. The vast number of EOCs and their occurrence in complex and variable mixtures is a major challenge for monitoring, risk assessment and management and is beyond the scope of target screening. Thus, novel approaches are needed to characterize these mixtures and identify unknown EOCs including transformation products and natural background. In this study, we propose a novel workflow for unravelling pollution patterns along a river course by identifying longitudinal dynamics of pollutant groups, entry pathways and the fate of EOCs along the river course using non-target screening by LC-HRMS and cluster analysis. Sixteen grab samples were taken along the 42 km- long course of the Holtmee River (Saxony-Anhalt, Germany), whereas the first sampling in the national park marked a reference point for pristine conditions. Chemical screening was performed on an UltiMate 3000 LC system (Thermo Scientific) coupled to a high-performance quadrupole hybrid - Orbitrap MS (QExactiveTM Plus, Thermo Scientific) with a heated electrospray ionization source. MS/MS analysis was performed in a full scan experiment (100-1000 m/z) at a nominal resolving power of 140,000 at m/z 200. Peak extraction including peak picking, gap filling, componentization and target annotation was implemented in R. Cluster analysis was performed using the R package ‘kmcl’. Four clusters were suggested for the data set representing A: EOCs from treated wastewater input of the two wastewater treatment plants (WWTP), B: EOCs specific for first WWTP due to specific local emissions, C: EOCs from diffuse (i.e., agricultural and urban surface run-off) and small point-source input (e.g., rain sewers and creeks) and D: low continuous background signals. The identified patterns gave insights into the spatial dynamics of complex chemical mixtures along a river course, highlighting differences in point-sources and areas governed by diffuse input and identifying points of complex mixtures of EOCs (e.g., first WWTP). Deeper investigation including structure elucidation will resolve the origin of non-target signals in these clusters. The proposed workflow proved to be a fast method for unravelling pollution patterns in non-target HRMS data and may also applied to study other longitudinal data such as temporal dynamics in pollution at hotspots and comparison of treatment and transformation processes.

20
Tracing sewage-derived contaminants from mainland towards the ocean by high resolution mass spectrometry
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The Ganges is the ultimate sink of most of the organic synthetic compounds produced and consumed by humans. Among the different pollution sources affecting this environment, discharge of treated and untreated sewage from mainland is of high relevance due to its continuous input, high volume and poor efficiency of conventional wastewater treatment plants (WWTP) to remove many potentially harmful substances. Even after dilution, some of these contaminants may still be detected at low concentrations (ppt-pb level), especially in coastal waters, and their effects over marine biota are still widely unknown. This work focused on identifying a wide range of polar and semipolar chemicals that can be detected in river water. Similar to river water, elevated concentrations of PPCPs and ASWs in groundwater can be interpreted as a consequence of inefficient wastewater management in the basin, which pose a concern for human exposure.

21
Pharmaceuticals, personal care products (PPCPs), and artificial sweeteners (ASWs) in river and groundwater from the Ganges River Basin, India
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Pharmaceuticals and personal care products (PPCPs) and artificial sweeteners (ASWs) are environmental contaminants of emerging concern. In this study, we investigated the occurrence and distribution of 15 pharmaceuticals and personal care products (PPCPs) and five artificial sweeteners (ASWs) in surface and groundwater of the Ganges River Basin in India. The Ganges River Basin is the largest river basin in India and home of about 7% of the total global population. PPCPs and ASWs were ubiquitously present in the river and groundwater. Most frequently detected compounds were caffeine, DEET, ketoprofen, cycloheximide and sacurlose. In this study, we measured concentrations of all PPCPs and ASWs in river and groundwater of the Ganges River Basin in India. The Ganges River Basin is the largest river basin in India and home of about 7% of the total global population. PPCPs and ASWs were ubiquitously present in the river and groundwater. Most frequently detected compounds were caffeine, DEET, ketoprofen, cycloheximide and sacurlose. Except caffeine and DEET, concentrations of other PPCPs and ASWs in surface and groundwater of the Ganges River Basin were lower but on same order as detected in the river water. Similar to river water, elevated concentrations of PPCPs and ASWs in groundwater were detected in middle and lower reaches along the Ganges River. PPCPs and ASWs were found to be lower in some of the developed countries, still, their instantaneous loads in the Ganges River were comparable to those in rivers from developed countries. The presence of PPCPs and ASWs in the surface and groundwater can be interpreted as a consequence of inefficient wastewater treatment processes in the basin, which pose a concern for human exposure.

22
Data-dependent fragment ion search for detection of sartans and related compounds in wastewater and surface water
B. Zoppa, IDAEA-CSIC / Environmental Chemistry; M. López de Alda, Institute of Environmental Assessment and Water Research-IDAEA-CSIC / Department of Environmental Chemistry; D. Barcelo, IQAB-CSIC / Department of Environmental Chemistry

EOCs from treated wastewater input of the two wastewater treatment plants (WWTP), B: EOCs specific for first WWTP due to specific local emissions, C: EOCs from diffuse (i.e., agricultural and urban surface run-off) and small point-source input (e.g., rain sewers and creeks) and D: low continuous background signals. The identified patterns gave insights into the spatial dynamics of complex chemical mixtures along a river course, highlighting differences in point-sources and areas governed by diffuse input and identifying points of complex mixtures of EOCs (e.g., first WWTP). Deeper investigation including structure elucidation will resolve the origin of non-target signals in these clusters. The proposed workflow proved to be a fast method for unravelling pollution patterns in non-target HRMS data and may also applied to study other longitudinal data such as temporal dynamics in pollution at hotspots and comparison of treatment and transformation processes.

23
HR-MS non-target analysis for transformation products of emerging organic contaminants in waste water fractions pre-screened by ELISA

High-Resolution Mass Spectrometry has its benefits but still wastewater samples challenge the analyst on the quest for "unknowns", metabolites and transformation products of emerging organic contaminants (EOCs). Their detection requires non-target analysis which involves not only costly instrumentation but also scientists with the time to plough through the enormous amount of data collected. An approach is presented using antibodies as selectors to pre-screen fractions of an...
HPLC run for “binding” in order to detect hitherto unknown but structurally related compounds. Carbamazepine (CBZ), an anti-convulsant and anti-depressant, sulfamethoxazole (SMX), an antimicrobial for humans, and estrone (E1), a hormone and estradiol metabolite have been studied by this LC-ELISA approach. Immunoassays had been developed for all compounds but overestimations of wastewater concentrations were frequent, with CBZ even at a constant level (+30%) that did not result from the consideration of reactivity to CBZ-10,11-epoxide (ca. 70%) or 2-hydroxy-CBZ (14%). Fractions from HPLC runs of pre-concentrated wastewater samples were collected into a 96-well glass plate in small aliquots, with fractions adapted to the desired resolution along the run. One plate is sufficient to collect a 30–40 min. run. The fractions were evaporated to dryness under a gentle stream of nitrogen. Before ELISA analysis, all fractions were dissolved in 100 µl of a loading buffer (TOF-SPE-MS) applied on a specifically “positive” fraction revealed an exact mass of m/z = 389.168 and a chlorine pattern. The compound is cetirizine, an antihistaminic. It appeared in our samples from spring on and was responsible for 20% of the overestimation we found initially with the CBZ antibody. The LC-ELISA for SMX displayed a series of unidentified peaks in the ELISAgram. Careful analysis of the fractions led to the identification of N4-acetyl-sulfamethoxazole, an SMX metabolite which is present in the samples. With estrone, interferences by polar matrix compounds eluting early could be identified.

24 Designing a risk based monitoring program for groundwater sources for drinking water production – based on target and suspect screening combined with clustering techniques
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Drinking water utilities heavily invest in monitoring occurrence of chemicals in drinking water sources and produced drinking water. Worldwide, drinking water regulation prescribes drinking water limits for a limited number of chemicals, the EU Drinking Water Directive (EU DWD) for example lists drinking water limits for 26 organic and inorganic chemical parameters. However, most drinking water utilities monitor a broad set of parent chemicals and their transformation products, using both target, non-target and bioanalytical methods. The EU DWD stimulates that drinking water monitoring is performed in a more flexible way, provided protection of public health is ensured. Compared to surface water, groundwater is less intensively studied and monitored. However, groundwater can be highly influenced, by anthropogenic activities related to the land-use above the groundwater, by infiltrating surface water, by historical contamination as well as by activities in the sub-soil. The susceptibility of the groundwater aquifers to these pressures depends on soil type and groundwater hydrology. Chemical properties such as persistence and mobility and their retardation during groundwater flow are reflected in the spatio-temporal patterns of the chemicals. Treatment technology applied, such as filtration and sorption techniques, determines removal efficiencies during drinking water production for specific compounds. Water utility Vitens services drinking water in a large area in the Netherlands, mostly using groundwater as a source. Their set of chemical parameters in the monitoring program tripled in the last decade. The water utility aims to prioritize their measured chemicals and develop a tailored risk-based monitoring program. We propose that the EU DWD is the step in developing a risk-based monitoring program for all 133 supply zones involved, mostly consisting of groundwater. We use both target and non-target/suspect monitoring data and well characteristics. We use clustering techniques combined with prioritization techniques including substance properties and in vitro as well as in vivo toxicity information. We analyse full scale removal efficiencies by the treatment technologies applied. Finally we propose a risk based monitoring program.

Wildlife ecotoxicology: laboratory dosing studies to field population assessments (I)

25 An interspecies correlation model to predict acute dermal toxicity of plant protection products to terrestrial life stages of amphibians
L. Welge, P. Janz, BASF SE, Crop Protection - Ecotoxicology; P. Sowig, Bayer CropScience / Ecotoxicology

In this presentation, a model to predict acute dermal toxicity of plant protection products to terrestrial amphibian life stages from (regulatory) fish data will be presented. By combining existing concepts, including interspecies correlation estimation (ICE), allometric relations, lethal body burden (LBB) and bioconcentration modelling, an equation was derived that predicts the amphibian median lethal dermal dose (LD₅₀) from standard acute toxicity values (96-h LC₅₀) for fish and bioconcentration factors (BCF) in fish. Where possible, fish BCF values were correlated to amphibian BCF and to parent compound. Then, BCF values were adjusted to an exposure duration of 96 h, in case steady state took longer to be achieved. The derived correlation equation is based on 32 LD₅₀ values from acute dermal toxicity experiments with 15 different species of anuran amphibians, comprising 15 different plant protection products. The developed ICE model can be used in a screening approach to estimate the acute risk to amphibian terrestrial life stages from dermal exposures to plant protection products with organic active substances. Applying this method has the potential to reduce unnecessary testing of vertebrates.

26 Overview of the EFSA Scientific Opinion on the state of the science on pesticide risk assessment for amphibians and reptiles
S. Pieper, German Federal Environment Agency (UBA) / Plant Protection Products; P. Adriaanse, Alterra Wageningen University and Research Centre; A. Albrecht, Agroscope / Ecotoxicology; C. Berg, Uppsala universitet, Dept. of Environmental Toxicology / Dept of Environmental Toxicology; P. Berny, VETAGRO-SUP / Toxicology; K. Machera, Benaki Phytopathological Institute / Department of Pesticides Control & Phytopharmacy, Athens, Greece; M. Ortiz Santaliestra, Institute for Game and Wildlife Research (IREC) UCLM-CSIC-JCCM; C. Topping, University of Aarhus; S.M. Weir, Queens University of Charlotte / Biology; F. Streissl, EFSA / Pesticides Unit; R. Smith, School of Applied Sciences, University of Huddersfield

Concerns have been raised that the current risk assessment schemes may not sufficiently cover the risk for amphibians and reptiles exposed to intended uses of plant protection products (PPP). To address these concerns, the European Food Safety Authority (EFSA) has published a Scientific Opinion on the state of the science on pesticide risk assessment for amphibians and reptiles. EFSA Journal 2017; 329 pp. doi:10.2903/j.efsa.20YY. [2] EC. 2009. Regulation (EC) No 1107/2009 of the European Parliament and of the Council of 21 October 2009 concerning the placing of plant protection products on the market. OJ L 39/1

27 Ecotoxicological assessment of Caretta caretta (Linnaeus, 1758) in the Mediterranean Sea using an integrated non-invasive-protocol
S. Casini, University of Siena / Scienze Fisiche della Terra e dell’Ambiente; I. Caliani, M. Giannetti, L. Marsili, S. Maltese, D. Coppola, N. Bianchi, T. Campani, University of Siena / Department of Physical, Earth and Environmental Sciences; S. Ancora, University of Siena / Physical sciences, Earth and environment; M. Fossi, University of Siena / Department of Physical Sciences, Earth and Environment Caretta caretta is the most common sea turtle in the Mediterranean Sea. The IUCN assessment for this carnivorous long-lived reptile underlines the lack of information regarding pollution and pathogens and indicates as a priority efforts to investigate and reduce the impacts of these threats. Up to now very few studies were conducted investigating biological endogenous potential biomarkers on C. caretta. The aim of our study was to conduct the first ecotoxicological assessment of this species in the Mediterranean sea using a non-invasive integrated methodology. We set up and applied a monitoring protocol which also includes endpoints, such as CYP1A, Lipid peroxidation, ENA assay, B esterases, never investigated before in this species. Seventy-five loggerhead turtles were sampled in a prospective way in B. Lampedusa Rescue Area free-ranging along the Spanish coasts. Blood, skin and carapace samples were used to test biomarker responses and contaminant (OCs, PAHs, Pb, Cd, Hg) levels. We measured biomarkers of exposure to lipophilic contaminants (CYP1A in skin biopsies), biomarkers of indirect and direct effects investigating neurotoxic (esterases inhibition) and estrogenic (vitellogenin) effects, oxidative stress (lipid peroxidation), genotoxicity (Comet and ENA assays) and liver damage (Gamma Glutamyl Transferase). Elaboration of experimental results was carried out taking also into consideration different age classes of the specimens. Among the main results obtained we should underline the statistically significant correlation between
carcinogenic PAHs in blood and DNA fragmentation as well as between Cd in carapace and GGT in plasma. We measured a very sharp band with a molecular weight of 59 kDa in skin sample that can be attributed to CYPIA1, never investigated earlier in this species. We also evidenced as the youngest animals showed significantly higher DNA fragmentations, BChE inhibition and increase of GGT; these alterations can be potentially related to their coastal habits. Older specimens showed the highest levels of erythrocyte nuclear abnormalities which may indicate a long term toxicological stress. This study contributed to expand the knowledge about the ecotoxicology of C. caretta in the Mediterranean, the non-invasive protocol could also be applied to other marine ecosystems and other sea turtle species, and implemented with new endpoints in the near future.

28 Sucking claws or hunting seals - consequences to walrus health
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The walrus (Odobenus rosmarus) is an ice-associated marine mammal with distinct feeding habits. Concentrations of the main chlorinated pollutants, namely polychlorinated biphenyls (PCBs) and chlordanes, in walruses that likely feed on seals are very high - similar to levels observed in polar bears, whereas pollutants concentrations in walruses feeding on benthos are lower. Although multiple studies have associated contaminant exposure to adverse health effects in polar bears and other marine mammals with low contaminant exposure, there are, to our knowledge, no studies to date investigating effects of pollutants in walruses. The goal of our study was to investigate contaminant and pathogen exposure and endocrine disruption in walruses feeding at different trophic levels. Samples from adult male walruses (n=39) were collected from Svalbard, Norway. Stable isotope values determined in seven body compartments indicated that all of the walruses in this study fed at a low trophic level. However, concentrations of blubber lipophilic compounds showed very high individual variation. Concentrations of chlorinated compounds have decreased since they were last studied in walruses sampled, in the same area as the current study, during 2002-2004. Plasma PFAS concentrations varied less between individuals. δ15N values in red blood cells and in other marine mammal species, respectively. HUHAH. HUHAH. HUHAH. HUHAH.

30 Egg overspray with herbicides and fungicides reduces chick survival in red-legged partridges
M. Ortiz Santaliestra, Institute for Game and Wildlife Research (IREC) UCLM-CSIC-JCCM; V. Alcaide, IRIAF / Centro de Investigación Agroambiental El Chaparrillo; R. Mateo, IREC-CSIC - UCLM / Grupo de Toxicología de Fauna Silvestre; F. Mougeot, IREC

Toxicity characterization in pesticide risk assessment for birds is derived from oral exposure of adults. However, for ground-nesting species, a temporal and spatial overlap of egg laying and incubation with pesticide applications may result in direct exposure of the eggs. Using formulations commonly applied to cereal crops in spring, we conducted two experiments in 2016. Likewise, potential according to field studies concluded that 2,4-D exposure during the oviposition period and effect occurrence need to be considered when monitoring pesticide impacts on avian reproductive output. Financed by the Spanish Ministry of Economy and Competitiveness (Project ref. CGL2016-75278-R)

Biocides and Veterinary Medicines: latest developments in regulatory risk assessment, research and monitoring

31 Regulatory improvement in the assessment of environmental risks from veterinary medicines: a European Perspective
J. Weeks, Joint Nature Conservation Committee

This presentation will focus on the developments in the environmental risk assessment of veterinary medicines from a European regulatory perspective. There is a need to continuously develop and react to changing requirements to perform more sophisticated, quantitative or robust environmental risk assessments for veterinary medicine products. The centralised approval of new veterinary medicines; a European Perspective
32 Risk of veterinary medicines to plants: Reflections for an updated approach. R. Carapeto Garcia, Spanish Medicines Agency / Veterinary medicines; A. Haro Castuera, Spanish Medicines Agency / Veterinary Medicines Department; G. Cotí Ruiz, C. Rubio Montejano, Spanish Medicines Agency / Department of Veterinary Medicines

In an Environmental Risk Assessment (ERA) the General Protections Goals need to be translated into Operational Protection Goals in order to achieve efficient and robust ERAs. Not doing so hinders the process of Risk Management in those cases where a risk is identified. In the current regulatory framework of ERA of Veterinary Medicines Protection of ecosystems (“Protection of ecosystems”) is not translated into Operational Protection Goals. Hence, when risks are found it is complicated to manage or mitigate such risks. In the taxonomic level of “terrestrial plants” some VMPs have shown different levels of risks. From the Risk Assessor perspective it is difficult to deal with these risks, partly due to the lack of guidance on Operational Protection Goals. Here we analyze a proposal of use of regulatory authorities as part of the drug approval process. Revalor-XR is an extended-release implant for use in cattle fed in confinement (steers and heifers) that contains trenbolone acetate and 17β-estradiol (17β-E2) as active pharmaceutical ingredients (APhIs). Both APhIs are metabolized in situ resulting in the excretion of 17β-trenbolone (17β-TB), 17α-trenbolone (17α-TB), trenbolone (TBO), 17β-E2, 17α-estradiol (17α-E2), and estroline (E1). The similarity in chemical structures and modes of the environmental fate properties among 17β-TB, 17α-TB, TBO, and that among 17β-E2, 17α-E2, and E1 promote the use of surrogate compounds to represent the trenbolone compounds and the estradiol compounds in the environmental assessment. Data on the individual compounds were collected from various laboratory studies and literature sources, aggregated to generate representative values for the surrogate compounds to characterize their environmental fate, and used for exposure assessments at field, field-ask and watershed-scales. Nine exposure pathways were evaluated at the feedlot and field scale, allowing for elimination of insignificant pathways for the watershed-scale modeling, which considered the major exposure pathways and was conducted for two representative watersheds, one in Texas and one in Iowa, using the U.S. EPA’s BASINS/HSPF model. The outputs of the modeling efforts resulted in Predicted Environmental Concentrations (PECs) for the surrogate compounds for individual as well as aggregated exposure pathways. The effects assessment was focused on potential reproductive impacts to fish from chronic exposure, which is the most sensitive ecological endpoint for these compounds, and generated Predicted No-Effect Concentrations (PNECs) for 17β-TB, 17α-TB, 17α-E2, and 17β-E2. Risk characterization involved comparison of the PECs for the surrogate compounds to the PNECs of the individual compounds. The assessment at the watershed scale demonstrated that it is highly unlikely that the compounds associated with Revalor-XR would have any significant environmental impacts when used according to the Revalor-XR label. The environmental assessment supported a Finding of No Significant Impact by the U.S. Food and Drug Administration from the use of Revalor-XR in beef steers and steers in the US.

33 Innovative environmental assessment of a veterinary medicinal product: watershed-level impacts of trenbolone acetate and 17β-estradiol J.P. Staveley, Q. Ma, J. M. Exponent, C. Celly, Intervet Inc. d/b/a Merck Animal Health; G. Scheef, MSD Animal Health Innovation GMBH / Preclinical Development

Environmental assessments of pharmaceuticals are required by regulatory authorities as part of the drug approval process. Revalor-XR is an extended-release implant for use in cattle fed in confinement (steers and heifers) that contains trenbolone acetate and 17β-estradiol (17β-E2) as active pharmaceutical ingredients (APhIs). Both APhIs are metabolized in situ resulting in the excretion of 17β-trenbolone (17β-TB), 17α-trenbolone (17α-TB), trenbolone (TBO), 17β-E2, 17α-estradiol (17α-E2), and estroline (E1). The similarity in chemical structures and modes of the environmental fate properties among 17β-TB, 17α-TB, TBO, and that among 17β-E2, 17α-E2, and E1 promote the use of surrogate compounds to represent the trenbolone compounds and the estradiol compounds in the environmental assessment. Data on the individual compounds were collected from various laboratory studies and literature sources, aggregated to generate representative values for the surrogate compounds to characterize their environmental fate, and used for exposure assessments at field, field-ask and watershed-scales. Nine exposure pathways were evaluated at the feedlot and field scale, allowing for elimination of insignificant pathways for the watershed-scale modeling, which considered the major exposure pathways and was conducted for two representative watersheds, one in Texas and one in Iowa, using the U.S. EPA’s BASINS/HSPF model. The outputs of the modeling efforts resulted in Predicted Environmental Concentrations (PECs) for the surrogate compounds for individual as well as aggregated exposure pathways. The effects assessment was focused on potential reproductive impacts to fish from chronic exposure, which is the most sensitive ecological endpoint for these compounds, and generated Predicted No-Effect Concentrations (PNECs) for 17β-TB, 17α-TB, 17α-E2, and 17β-E2. Risk characterization involved comparison of the PECs for the surrogate compounds to the PNECs of the individual compounds. The assessment at the watershed scale demonstrated that it is highly unlikely that the compounds associated with Revalor-XR would have any significant environmental impacts when used according to the Revalor-XR label. The environmental assessment supported a Finding of No Significant Impact by the U.S. Food and Drug Administration from the use of Revalor-XR in beef steers and steers in the US.

34 How can mesocosm studies increase realism in risk assessment of biocides and veterinary medicines? L. Dören, ERM / Product Stewardship; U. Hommen, Fraunhofer IME; P. Eb, R. Ovesen, Danish Environmental Protection Agency; H. Bakgaard, Copenhagen Fur

Bactericides are regulated in EU by the BPR [1]. To evaluate if an active substance (a.i.) or product may be authorised, an assessment of the environmental exposure is required. For insecticides used in stables an Emission Scenario Document (ESD) [2] is used covering application methods and a range of animal categories. The ESD does not cover biocides used in a manure stall. A scenario has therefore been developed, where emission of a.i. from manure is calculated based on either amount applied or measured concentration in straw. Default values have been established from regulation and general practice in manure in the Nordic countries, where Denmark has the highest production of mink in Europe [3]. Each breeding animal is kept individually in one cage to be treated at the start of the season. Mother and cub stay together in one cage and are separated into pairs after lactation, where all cages are treated. Each mother will bear 5.55 cubs/year according to Danish regulation [4]. The number of “breeding females” (BF) is 1 mother + 5.55 cubs. The number of nest boxes that is treated/BF may be calculated as follows: 1 animal/nest box before separation and 6.55/2 animals (3.275) nest boxes after separation. In Europe it is prohibited to discharge waste from stables to public sewer. Emission is therefore only expected to be to agricultural land. Emission of manure/straw may be from up to 50 BF per hectare (ha) per year based on regulation in the Nordic countries. Emission according to application pattern: Y = Qmax x Fmax x V = Napp before sep x 3.275 x Napp after sep x 18 x 10^2 (Eq. 1) Where Y is emission of a.i. in kg/ha x year, Qmax is amount of product/nest box in g, Fmax is concentration of a.i. in the product in kg/g, Napp before sep is number of treatments before separation of adults and cubs, Napp after sep is number of treatments after separation of adults and cubs and B is amount of straw/manure that may be applied (a.i. in kg/ha) before separation of adults and cubs. Emission from manure/straw is 10-15 kg/animal/year according to Copenhagen Fur. The emission based on Nordic countries regulations and information from Copenhagen Fur on amount of straw used per BF is 50 BF per ha x 15 kg straw per BF = 750 kg straw per BF per ha. Predicted Environmental Concentration in soil may be calculated according to Volume IV Part B [5].

35 Estimation of insecticides in mink farms R.G. Ovesen, Danish Environmental Protection Age; H. Bakgaard, Copenhagen Fur

Biocides are regulated in EU by the BPR [1]. To evaluate if an active substance (a.i.) or product may be authorised, an assessment of the environmental exposure is required. For insecticides used in stables an Emission Scenario Document (ESD) [2] is used covering application methods and a range of animal categories. The ESD does not cover biocides used in a manure stall. A scenario has therefore been developed, where emission of a.i. from manure is calculated based on either amount applied or measured concentration in straw. Default values have been established from regulation and general practice in manure in the Nordic countries, where Denmark has the highest production of mink in Europe [3]. Each breeding animal is kept individually in one cage to be treated at the start of the season. Mother and cub stay together in one cage and are separated into pairs after lactation, where all cages are treated. Each mother will bear 5.55 cubs/year according to Danish regulation [4]. The number of “breeding females” (BF) is 1 mother + 5.55 cubs. The number of nest boxes that is treated/BF may be calculated as follows: 1 animal/nest box before separation and 6.55/2 animals (3.275) nest boxes after separation. In Europe it is prohibited to discharge waste from stables to public sewer. Emission is therefore only expected to be to agricultural land. Emission of manure/straw may be from up to 50 BF per hectare (ha) per year based on regulation in the Nordic countries. Emission according to application pattern: Y = Qmax x Fmax x V = Napp before sep x 3.275 x Napp after sep x 18 x 10^2 (Eq. 1) Where Y is emission of a.i. in kg/ha x year, Qmax is amount of product/nest box in g, Fmax is concentration of a.i. in the product in kg/g, Napp before sep is number of treatments before separation of adults and cubs, Napp after sep is number of treatments after separation of adults and cubs and B is amount of straw/manure that may be applied (a.i. in kg/ha) before separation of adults and cubs. Emission from manure/straw is 10-15 kg/animal/year according to Copenhagen Fur. The emission based on Nordic countries regulations and information from Copenhagen Fur on amount of straw used per BF is 50 BF per ha x 15 kg straw per BF = 750 kg straw per BF per ha. Predicted Environmental Concentration in soil may be calculated according to Volume IV Part B [5].
that washing and cleaning agents are important sources for preservatives such as Bt and Otf, while triclosan was apparently mainly emitted through personal care products. The mosquito repellent substances DEET and icaridine were found throughout the whole year, with highest emissions in summer and autumn. C12-benzalkonium chloride concentrations were associated with the inventoried disinfectants. Material preservatives such as terbutryn, diuron, tebuconazole or cationic silver were also detected. As these were not listed on the inventoried products, emission via treated materials such as paint, render, seals or textiles seems likely. We were able to show that biocidal active substances are emitted from the inside of households in considerable concentrations. Those emissions are not only due to biocidal products but also washing and cleaning agents, personal care products and preserved materials. For this reason, measures should not only tackle biocidal products but also its concentration in wastewater. The environment as a reactor determining fate and toxicity of nanomaterials

37 Comparative multi-generation study of long-term effects of pristine and wastewater-borne silver and titanium dioxide nanoparticles on reproduction in Daphnia magna

S. Hartmann, University of Siegen, Institute of Biology / Department of Chemistry and Biology; R. Louch, University of Manchester; R. Zeuner, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Bioaccumulation and Animal Metabolism; C. Schlechtriem, Fraunhofer IME / Department Bioaccumulation and Animal Metabolism; K. Witte, University of Siegen / Department of Chemistry and Biology

Manufactured nanomaterials (MNMs) and especially Ag- and TiO2-NPs are produced in daily used products such as cosmetics, clothing and in medical supplies. After passing wastewater treatment plants these MNMs reach the aquatic environment and can accumulate in the aquatic ecosystem and cause toxicity to aquatic organisms. To assess the risk potential of these NPs to aquatic invertebrates under more realistic circumstances, we investigated and compared possible effects of pristine Ag-NPs and TiO2 NPs with those after passing a model wastewater treatment plant on the reproductive success (number of offspring), mortality and body size of adult daphnia as endpoints in up to six generations. We exposed daphnia to: (i) pristine Ag-NPs (NM300K) and TiO2-NPs (NM105) or (ii) wastewater borne Ag- and TiO2-Nanoparticles from effluent from the model WWTP. The first generation of daphnia was exposed to four concentrations of Ag-NPs (nominal: 1.25 µg/L, 2.5 µg/L, 5.0 µg/L and 10.0 µg/L), to solvent control (NM100K DIS), or to three concentrations of TiO2-NPs (nominal: 25 µg/L, 50 µg/L, 100 µg/L) in line with the OECD guideline No. 211. Each generation was exposed for 21 days and started with the third brood from the previous one. In all six generations the exposure with pristine Ag-NPs (NM300K) for 21 days caused a significant reduction in the mean number of offspring in daphnia compared to the control. However, wastewater-borne Ag-NPs had no effects on reproduction in any generation in the in vivo test and the body length of daphnia was significantly larger at 5 µg/L in generation F2 and at 2.5 µg/L in generation F3 compared to the control. In the wastewater-borne Ag-NP treatment the adults’ body length was significantly larger at 2.5 µg/L. Thus, adult’s body length showed no consistent pattern between both scenarios. When passing WWTPs most Ag-NPs might be transformed and enter the aquatic environment as silver sulfide. That may be the reason for the lower toxicity than compared to other forms of Ag-NPs. Our results provide a first, direct comparison between the toxicity of pristine Ag-NPs and TiO2-NPs with those from WWTP. To our knowledge, the present study is the first one showing that Ag-NPs from a wastewater treatment plant had a minor and no chronic toxicity to Daphnia magna. The used experimental approach allows a realistic risk assessment of Ag-NPs and TiO2-NPs for the aquatic environment. The experiment with TiO2-NPs are in progress.

38 Development of a rapid screen to assess bioaccumulation potential: from ex vivo to in vivo using pristine and aged nanomaterials in fish

A. Clark, School of Biological Sciences, Plymouth University; D. Boyle, Plymouth University; R. Hardy, University of Plymouth

Bioaccumulation is one of the key triggers of concern for environmental risk assessment. Among the factors that may influence the aquatic bioavailability of a nanomaterial, the size and composition of the nanoparticles, and the exposure route of the organisms to the material are of particular relevance. In the present study, we aimed to develop an ex vivo method that allows rapid screening of the bioavailability and potential of nanoparticles to pass the gut epithelium, which may be used to assess the potential of these materials to exert toxic effects on aquatic organisms. The method can be used to rapidly screen the bioavailability of Ag NPs and Ag2S NPs. However, if the data are ranked in the mid and hind intestine by total Ag accumulation, the gut sac does not directly predict in vivo accumulation.

39 Fate and Effect of Wastewater Borne Manufactured Nanomaterials on the Aquatic Food Chain

R. Zeuner, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Bioaccumulation and Animal Metabolism; B. Knopf, Fraunhofer Institute for Biomedical Milieu- and Environmental Toxicology IME / Department of Analytical Toxicology; S. Hartmann, Fraunhofer IME / Department Bioaccumulation and Animal Metabolism; K. Witte, University of Siegen / Department of Chemistry and Biology

Manufactured nanomaterials (MNMs) are widely used in various applications and commercial products, e.g. textiles, sunscreens, paints, cosmetics. Even though MNMs are mostly removed during wastewater treatment, the remaining and mostly transformed MNMs in the treated effluents may significantly increase the toxicity for aquatic organisms due to their modification during the WWT. The impact of wastewater-borne MNMs (TiO2 and Ag MNMs) on different trophic levels and their bioaccumulation within a relevant food chain (algae-Daphnia-fish) have been investigated with innovative analytical and experimental approaches. Several model WWTPs were conducted according to OECD guideline 304A. The collected effluents were used to perform acute and chronic tests with Daphnia magna and Onchorhynchus mykiss according to the OECD guidelines 202, 211, 215 and 305. Animals were exposed to: (i) effluent from model WWTPs contaminated with MNMs, (ii) untransformed effluent, manually spiked with MNMs and (iii) dilution water enriched with pristine MNMs. Tissue samples of the different test organisms were analyzed for changes in the levels of several biochemical markers (lipid peroxidation, DNA damage, lactate dehydrogenase (LDH), superoxide dismutase (SOD), catalase (CAT) and glutathione-S-transferase (GST)). Furthermore, uptake and elimination kinetics of the MNMs were investigated by quantitative ICP-MS and ICP-OES analysis. No chronic effects were found in D. magna after exposure to effluents with transformed AgNPs. However, when supplemented into untransformed effluents with AgNPs in the aquatic food chain, the algae-Daphnia-fish system showed a clear increase in the intoxicity of aquatic organisms. For nano-TiO2, no effects on the reproduction of D. magna could be shown at environmentally relevant concentrations. Only after chronic exposure to the very high concentrations of 5 mg/L and 10 mg/L significant effects could be shown. Neither nano-Ag nor nano-TiO2 showed an effect on the growth of juvenile rainbow trout after 28 days of exposure. The analysis of biochemical biomarkers showed that several effects induced by chronic exposure to MNMs were observed. However, no general effect pattern could be identified. Total MNM levels were measured in several tissue samples in D. magna and O. mykiss following exposure via the water or food. Pristine nanomaterials showed a significantly higher uptake into the test organisms compared to supplemented and treated WWTP effluents.
was established and combined with a battery of ecotoxicological assays and characterization techniques. The system contained activated sludge and was operated as a pre-denitrification system fed with synthetic wastewater spiked daily with 10 µg Ag NPs/L (PVP coated, 25 nm, nanoComposix) and 100 µg TiO$_2$ NPs/L (nominal primary size of 5 nm, NM-101, JRC) over a period of 5 weeks. During that period the effluents were collected weekly and the excess sludge was stored for the evaluation of temporal trends. Some samples of the sludge were collected weekly and analyzed by sequential filtration and ICP-MS to determine the partitioning of NPs and their transformation products. Transmission electron microscopy and sp-ICP-MS were performed on selected samples. The effects of aged NPs were assessed using a battery of bioassays including freshwater and marine algae (growth inhibition and reactive oxygen species -ROS-) formation, and crustaceans, as well as in vitro models of relevance for NP toxicity assessment (RTgill-W1 cell line, effects on metabolic activity, epithelial integrity, ROS formation, gene expression). The extent of the observed effects was dependent on the organism exposed, with bottom feeding organisms and algae being more sensitive, while the in vitro model was a good tool for environmental samples. Furthermore, the biosolids generated from the lab-scale continuous system were used for terrestrial microcosm experiments, giving insight into the fate and potential accumulation in a model terrestrial system. Experimental data generated from the continuous-flow operation of the activated sludge system and the targeted batch experiments will be used to model the fate and the removal of NPs.

Advances in environmental risk assessment of oil spills and offshore oil & gas operations (I)

43 Optimization of Oil Spill Response Planning and Preparedness Using Spill Mitigation Impact Assessment (SIMA)


Oil spill emergency response plans (OSRP) are required as part of permitting for offshore operations. An OSRP typically includes risk assessment and detailed plans for responding to different types of oil spill accidents involving shipping, pipelines, platforms, and/or subsea wells. The owner/operator must demonstrate to state and federal regulatory authorities that the company’s OSRP for offshore exploration and production operations conform to all applicable regulations and international standards and practices, and further demonstrate that the necessary equipment and trained personnel are in place to respond quickly and effectively to an oil spill accident. In the event of an oil spill accident, the priorities for oil spill response (OSR) are to protect people, prevent or mitigate environmental damages, and prevent impact to affected communities. Spill Impact Mitigation Assessment (SIMA) is a science-based framework evolved from Net Environmental Benefits Analysis (NEBA) to broaden the focus from consideration of mitigation of ecological impact to include mitigation of socioeconomic and cultural impacts, as well. SIMA is a method for identifying and comparing the socioeconomic and -economic benefits of alternative OSR options, with the goal of selecting options that best mitigate the consequences of spilled oil and impose the lowest additional negative consequences on environmental and social resources. SIMA is both site- and spill- specific, and is particularly useful during the initial planning and preparedness period for oil and gas exploration and when new technologies and best practices emerge that need to be adopted into safety, health and environmental management programs. This paper describes a spill impact mitigation assessment framework using recent examples of OSRP work conducted for the Arctic Oil Spill Response Technology Joint Industry Project, American Petroleum Institute Gulf of Mexico Deepwater Project, and several companies working in tropical marine environments. The applicability of SIMA to marine resources and habitats is also discussed. The SIMA framework typically includes (a) assessment of initial impacts and potential consequences in the marine environment, and (b) effectiveness and consequences of deploying different spill response strategies.

44 Adapting the SIMA Process to Assess Offshore Decommissioning Options

T. Coolbaugh, ExxonMobil Research & Engineering; A. Aziz, ExxonMobil Upstream Research Company; P. Taylor, Petronia Consulting Limited; G. Coelho, Spoon Group Inc.

For several decades, the oil and gas industry has used the Net Environmental Benefit Analysis (NEBA) approach for oil spill response contingency planning. Recently, IP/EC/E-API/IOGP published guidelines on the implementation of NEBA, using a novel process known as Spill Impact Mitigation Assessment (SIMA) that directly addresses the potential consequences of spills and provides a more consistent and transparent approach. This paper describes SIMA and presents examples of OSRP work conducted for the Arctic Oil Spill Response Technology Joint Industry Project, American Petroleum Institute Gulf of Mexico Deepwater Project, and several companies working in tropical marine environments. The applicability of SIMA to marine resources and habitats is also discussed. The SIMA framework typically includes (a) assessment of initial impacts and potential consequences in the marine environment, and (b) effectiveness and consequences of deploying different spill response strategies.
drawbacks of each option, thereby developing response strategy. Similarly, oil and gas operators are faced with complex options for decommissioning offshore installations as part of their decommissioning plans. There is a need to evaluate these options in a scientifically-defensible and consistent manner, while adequately assessing risks that can be challenging to assess. An adapted SIMA process could be a valuable tool for fostering collaboration between operators, stakeholders and regulatory authorities in ensuring a transparent review of engineering studies and available decommissioning options in a meaningful way. This paper evaluates the use of the SIMA process in a hypothetical decommissioning of an offshore platform to determine if this process lends itself to this purpose. It describes the challenges encountered when trying to conduct this comparative risk assessment, originally intended for oil spill response, to the task of decommissioning. Finally, it proposes adaptations to the SIMA process that might enable its use in the future as a credible tool in assessing environmental risks for oil and gas decommissioning.

45 Detection and quantification of oil contamination in vegetated areas using hyperspectral remote sensing

G. Lassalle, ONERA; S. Dent, ONERA / Optics and Associated Technics; A. Credo, R. Hédaq, TOTAL SA / Environment; P. Borderies, ONERA / DEMR; G. Bertoni, INRA / Dynafor; D. Dubucq, TOTAL SA; A. Elger, University of Toulouse in / Ecodec

In the field of oil and gas production, there is a constant challenge in developing new techniques of oil detection for prospecting (natural seeps) and environmental monitoring purposes. Using airborne hyperspectral imaging (AHSI), it is possible to discriminate between sites affected by spills and those that are not. The objective of this presentation is to demonstrate the potential of this technique for assessing environmental risks deriving from chemical compositions. A pilot scale coastal in situ burning operation was performed during summer in a bay in western Greenland with a crude oil for testing burning efficiency and environmental exposure and effects. The studies were funded by the European Commission Horizon 2020 programme and the Government of Greenland.

46 A tool for tracking complex ecotoxicological effect data after large pollution events with use of the Deepwater Horizon oil spill as a case study

J. Beyer, NIVA - Norwegian Institute for Water Research; H.C. Tramum, T. Bakke, Norwegian Institute for Water Research; P.V. Hodson, Queens University / School of Environmental Studies; T.K. Collier, Delta Independent Science Board Norwegian Research Council

The Deepwater Horizon Oil Spill (DWHOS) in 2010 is the largest and most studied accidental marine oil spill in history. More than 100 new research studies concerning the effects of the DWHOS have been published each year since 2011. Key issues investigated include the behaviour and fate of oil in deep spills, the effects on dispersed fractions, microbial oil degradation, oil-affected marine snow formation, oil impacts on deep water corals, seafood quality and safety, oil contaminants effects on fish, birds and marine mammals, effects of combined stressors on species and habitats, and habitat and ecosystem recovery processes. To keep order in this flow of new knowledge is an important albeit challenging task. It is essential that the lessons of DWHOS are applied globally to the next big marine oil spill, wherever it may happen. In this presentation, we demonstrate a convenient tool for keeping track of the large amount of ecotoxicological data and knowledge that typically emerges from research and monitoring after marine pollution disasters, using the DWHOS as a case study. In addition, we provide a summary of the new insights about oil spill effects on marine ecosystems that have been gained from the DWHOS research, and identify some key knowledge gaps still remaining. The presentation will update a comprehensive review about the environmental effects of DWHOS that we recently published in Marine Pollution Bulletin.

47 Oil spill combat and effects in the Arctic coastal environment; self-cleaning potential and in situ burning

S. Wegeberg, Aarhus University / Department of Bioscience; J. Fritt-Rasmussen, Aarhus University / Department of Bioscience - Arctic Environment; O. Gjerde-Hansen, M.R. Larsen, Greenland Institute of Natural Resources; K. Gustavson, Aarhus University / Department of Bioscience - Arctic Environment

What is the environmental effects of a beaches oil spill in the Arctic, how well will the shoreline potentially be able to self-clean and will combating the oil by in situ burning at the coast just do more harm to the communities in the tidal zone? To answer these questions, several studies have been performed at the west coast of Greenland in 2016 and 2017. One aim of the studies was then to support net environmental benefit analysis, NEBA, related to oil spill in Arctic waters. A NEBA is often performed to achieve the optimal environmental effect with respect to choice of oil spill combat methodology and biology at risk. Hence, a synthesis will be presented of following studies: 1) removal rate and ecotoxicological effects of oil smother on seaweed (Fucus distichus), an important organism of the communities in the coastal tidal zone; 2) self-cleaning potential of a coast line, including natural removal by seawater wash and physical degradation; and 3) effects on the tidal communities after combat of a beaching oil spill by in situ burning. Effects of oil smothering of the macroalgae Fucus distichus, which inhabit the intertidal zone of the coasts in the Arctic, as well as its self-cleaning potential by wash in sea were studied over a period of 2 weeks in a field test at Disko Island on the Greenland west coast. The当日 tiled resulted in that the tidal removal rates of correlation to different water cover regimes and air exposure times were obtained. The oil remain on the tiles were analysed for chemical compositions. A pilot scale costal in situ burning operation was performed during summer in a bay in western Greenland with a crude oil for testing burning efficiency and environmental exposure and effects. The studies were funded by the European Commission Horizon 2020 programme and the Government of Greenland.

48 How stable are our indices? - differentiating between sources in a weathering environment

S.M. Mudge, NILU - Norwegian Institute for Air Research / IMPACT

Crude oil contains hundreds of compounds and some of these are widely used to differentiate between different oils and products, especially in spill scenarios. Traditionally, we have developed chemical fingerprints based on a suite of compounds such as the steranes and terpanes although the concept of a “fingerprint” suggests this is static in time. However, it is also well known that these compounds are likely modified. Compared to differentiating between sources in a weathering process change the chemical signature and oil may have a different chemical composition to the original source oil. When we analyses such samples, we may need to ask if this is the same oil as the proposed source, or a different oil with a different signature that is also present. The steranes and terpanes contain several homologues and analysis of the chemical signature during the Deepwater Horizon Response clearly indicated that several of these compounds were not behaving conservatively and were degrading at a faster rate than anticipated given the exposure time. Comparisons with the actual oil released clearly identifies the compounds most likely to alter the environments where they degrade. In this case, the Louisiana marshes were clearly a site where biodegradation was significantly faster than expected. This was also true of the alkylated PAHs which had been used as source identifiers in previous spills such as the Exxon Valdez. The triaromatic steranes were also degrading at a significant rate while the oil was at sea and the exposure to UV light may have led to a relatively rapid abiotically transformation. When it comes to distinguishing between sources, less may be more! We need to select the compounds we include in our analyses with care since each question may need a different approach: if we want to know if the oil is weathering, we use a suite of compounds with differential properties appropriate to the environment of the spill. If we want to conduct source apportionment, we may need to choose the most recalcitrant of the compounds rather than all of them.

Fish model species in human and environmental toxicology (I)

49 Exposure to bisphenol S alters microRNA expression in male zebrafish (Danio rerio)

J. Lee, J. Ji, Yongin University

In response to the restriction of bisphenol A (BPA), bisphenol S (BPS) has been widely used in the manufacturing of polycarbonate plastics and epoxy resins as an alternative compound. BPS has been found to affect reproduction, development,
and immune system. Although microRNAs (miRNAs) play a crucial role in many metabolic activities, whether and how they are involved in the process of BPS-induced toxicity is largely unknown. BPS-induced changes in miRNAs and target gene expression in male zebrafish (Danio rerio) gonad, and the potential mechanism was investigated. Male zebrafish were exposed to 0, 5, and 50 μg/L BPS for 21 d. miRNA was isolated from the gonad pool and the expression profiles of 255 known zebrafish miRNAs were analyzed using Affymetrix microarrays. Quantitative real-time PCR was used to validate the expression of several miRNAs in the microarray data. The GO term analysis revealed that miRNAs significantly affected by BPS exposure were involved in hematopoiesis, lymphoid organ development, and immune system development. Among 14 miRNAs that were significantly regulated after exposure to 5 and 50 μg/L BPS, six miRNAs (miR-127, miR-138, miR-142a, miR-142b, miR-142c, and miR-196a) showed the opposite trend in BPS-induced toxicity via the interference with the aromatization process. The results of this study will provide novel insight into the epigenetic regulatory mechanisms of BPS-induced toxicity in male zebrafish.

50 Zebrafish as a model to investigate mechanisms of adverse metabolic and cardiovascular outcomes associated with elevated dietary selenium exposure

D.M. Jang, University of Saskatchewan / Toxicology Centre; C. Pettem, University of Saskatchewan - Toxicology Centre / Toxicology; J. Thomas, University of Saskatchewan Toxicology Centre; L.P. Weber, University of Saskatchewan / Veterinary Biomedical Sciences

A variety of in vitro and in vivo experiments indicate increased loading of the essential trace element selenium into aquatic ecosystems, where it poses an extreme toxicological hazard to fishes due to the narrow range between essentiality and toxicity. Although several studies have reported developmentally toxic effects in early life stages of fishes, fewer studies have investigated sublethal toxicological effects that may occur following dietary selenium exposure in adult fishes. Adult zebrafish were exposed to dietary selenium (Se) at sublethal (2 μg/L Se for 28 d) and environmentally relevant supraphysiological levels (3.4 – 28.8 μg/g) for 90 days. Swimming performance, O2 consumption and metabolic rates were determined using a swim tunnel respirometer. Cardiac function was assessed using high resolution (30 μm) ultrasound biomicroscopy. Whole-body energy stores (triglycerides and glycogen) and mRNA transcript abundance of selected genes of interest were determined. Compared to controls, adult zebrafish exposed to elevated dietary SeMet exhibited impaired swimming performance (lower fatigue velocity or Ucrit). This was associated with elevated basal metabolic rate and reduced aerobic scope, indicating impaired aerobic capacity. Triglycerides (the primary fuel for aerobic swimming) were elevated in a dose-dependent manner, which was associated with altered transcript abundance of several genes involved in lipid homeostasis. Ultrasonography revealed decreased cardiac output, which was associated with increased echodensity at the atrial-ventricular junction and reduced mRNA expression of the collagean, MMP2. These results suggest significant ecophysiologicological effects that may impair the fitness of fishes exposed to elevated dietary Se in contaminated ecosystems. From a comparative biomedical viewpoint, the zebrafish model leads to the identification of metabolic, energetic, and cardiovascular toxicities caused by excess dietary Se exposure, since similar responses following selenium over-supplementation have been reported in the human clinical toxicology literature. A proposed adverse outcome pathway (AOP) based on this study will be presented that links changes in gene expression to key events leading to adverse outcomes at the individual, and potentially population, levels of biological organization.

51 Toxicity and neurotoxicity profiling of sediments from Gulf of Bothnia with Danio rerio embryos

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Sediments are a well-known sunk for a large variety of organic pollutants that may cause distress to benthic and pelagic species in case of their remobilization to the water-phase. Risk assessment of complex mixtures may involve component-based approaches applying chemical analysis together with measured or predicted toxicity data of individual components and mixture risk modeling or whole mixture approaches using existing multi-chemical approaches (e.g., CPFs). This study focuses on the development of approaches combining both approaches applying chemical and bio-analytical tools for characterization for example of sediment contamination is expected to provide a more comprehensive picture of toxic risks to aquatic organisms. One of the most promising organisms for diagnostic in vivo testing of sediment may be zebrafish embryos (Danio rerio) being a versatile in vivo model suitable for high-throughput analysis while keeping several advantages of in vitro approaches (i.e., low-cost, sensitivity, short duration of the test). The fish embryo test (FET) with Danio rerio has been considered as a good surrogate for the acute toxicity fish test and was successfully used in several studies for the detection of toxicity and neurotoxicity in sediments samples. One of the major advantages of the FET with Danio rerio is the possibility to monitor several toxic endpoints including the modification of biochemical and molecular processes, which can be related to the exposure of specific pollutants. The present study provides a first attempt to integrate a diagnostic whole mixture assessment workflow based on in vivo toxicological profiling of Danio rerio after direct exposure to sediments from Gulf of Bothnia (Sweden) for 4 days. The objectives of the present study was (1) to validate a screening approach for sediment of samples (2) to offer a first in vivo toxicological profile of sediment from three different polluted sites from Gulf of Bothnia.

52 Proteomics based screening tool to detect molecular responses following aromatase inhibition

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Chemical exposure to endocrine disruptors can have adverse outcomes on organism health and function; however, the current reliance on end-points such as egg number, plasma VTG content and morphological changes to determine effects of endocrine disrupting chemicals has given rise to series of questions related to chemicals exhibiting similar effects but different mode-of-action (MoA). Improvements identified to the present study is the use of zebrafish to identify not only structural end-points but also potential biomarkers to determine chemical-induced adverse outcomes of ecological relevance in order to avoid unnecessary extensive testing.

53 Zebrafish embryos are able to conduct complex biotransformation processes and activation of chemicals

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Zebrafish embryos are proposed and partially already used as replacement for tests with (adult) animals conducted for human and environmental hazard assessment. Zebrafish have been considered to be more potent species which could interfere with prediction of adverse effects for safety assessment. The concern was provoked by anecdotal observation of the lack of activation for some compounds leading to a weak acute toxicity in fish embryos. However, internal concentration profiles of parent compounds suggest that fish embryos are principally capable for metabolic transformation of chemicals (Kühnert et al., 2013, Environ. Toxicol. Chem. 32, 1819–1827). In order to assess the biotransformation more systematically we studied the biotransformation of two pharmaceuticals (clofibrate, celecoxib). Overall similar transformation products could be observed in zebrafish embryo as known from human studies. Interestingly, the ratios of the different products in fish embryos seemed to be different from the ratio in humans. Biotransformation is of particular relevance for compounds that require (metabolic) activation to elicit their intended biological effect or where activation “accidentally” produces more toxic substances. Organophosphates (OP) can serve as a model compounds for a proof of concept for activation in fish embryos as in many cases activation is mediated by cytochrome P450 oxidation. Therefore, we studied the
activation of organophosphates by comparing the inhibition of acetylcholinesterase (ACHE) activity for the parent compound and the oxon-metabolite in homogenates and intact embryos. For homogenates only the oxon metabolites were able to provoke ACHE inhibition in a concentration dependent manner. In intact embryos inhibition was also found for the parent compounds. The inhibition EC50 was gradually reduced in later stages of embryos, if 24h exposure intervals for different stages were compared. The findings suggest that fish embryos are principally able to activate organophosphates and potentially also other compounds. A disadvantage for later embryonic stages may exhibit advanced biotransformation capacity.

54 Differing PM2.5 Filter Extraction Methods: Impact on Chemical and Toxicological Analyses C. Roep, Oregon State University / Department of Environmental and Molecular Toxicology; S. Simonich, Oregon State University / Depts of Chemistry and Environmental Molecular Toxicology; R.L. Tanguay, Oregon State University / Simnubber Aquatic Research Laboratory and the Environmental Molecular Toxicology

Toxicology research is essential to improve the understanding of the global public health burden of fine particulate matter (PM2.5) exposures. However, research groups use differing filter extraction methods to prepare PM2.5 and the potential toxicity bias from different extraction methods is rarely considered, possibly eliminating inter-laboratory comparisons and misrepresenting the toxic responses to PM2.5 constituents. To determine the impact of filter extraction methods on chemical constituent recovery and toxicity outcomes we took equal portions of a single high-volume PM2.5 filter sample collected in Riverside, CA. Each filter portion underwent a different extraction method (n=6) and recovered PM2.5 was then prepared for developmental toxicity testing by collecting the soluble fraction from DMSO extraction. Zebrashin (n=32/treatment) were treated with controls (DMSO, blank filter portions) and treatments (PM2.5 filter portions undergoing filter extraction) starting at 6 hours post fertilization. Aliquots of these PM2.5 solutions were used for chemical constituent analysis of polycyclic aromatic hydrocarbons (PAHs, n=120) and elements (n=20). Significant increases in mortality were observed for PM2.5 from 5 of the 6 filter extraction methods when compared to both the DMSO and blank filter controls. Combined mortality and morphological changes were significantly increased following PM2.5 treatment in all extraction methods compared to DMSO controls. Important, two of the methods showed significant mortality and morphological changes with blank filters when compared to DMSO controls. Chemical analysis is underway and differences in PM2.5 solutions between extraction methods will be investigated. Correlations between chemical components and developmental toxicity outcomes will identify constituents that are driving mortality and potentially altered during specific extraction procedures. This research highlights the toxicity bias due to PM2.5 filter extraction methods that must be considered when conducting research with complex ambient mixtures. Ultimately, this work identifies extraction procedures for use in this cost-effective surrogate to compare the inherent toxicity differences of PM2.5, and provides a path that will ultimately promote improved understanding of PM2.5-associated health effects.

Safeguard and Conservation of Cultural Heritage: the contribution of chemistry

55 Cultural Heritage and Climate Change: impact and adaptation C. Sabbioni, CNR-Istituto di Scienze dell’Atmosfera e del Clima Cultural heritage, which is a non-renewable resource, is a sector extremely complex for the diversity of materials, structures and systems. The access to citizens and visitors needs to be favoured, but at the same time, it is our responsibility to transmit this heritage we received from the past to the future generations. It is urgent to include cultural heritage in the value chain of sustainable development: the priority that faces the world today. Research on the threats that climate change will have on cultural heritage has been very limited until now and it has not yet generated policies designed to mitigate the impact and to develop preventive adaptation strategies. The presentation will be focused on future scenario on the effects of climate change on the vulnerability of cultural heritage at European level. Recommendations on the inclusion of cultural heritage in the national adaptation strategies and plans to climate change will also be discussed.

56 Nanotechnologies for the conservation and connected risks M.J. Mesquera, University of Cadiz Most products commonly employed in the restoration and conservation of cultural heritage materials have not been specifically developed to preserve such elements. In addition, they are plagued by limited performance and structural drawbacks such as low adhesion, poor penetration, and cracking. Another disadvantage is the requirement for most products to be dissolved in volatile organic compounds (VOCs), which produce environmental and human health risks in their use. In this lecture, I will review the most meaningful achievements of my group in this field. We have developed an innovative sol-gel route for preserving Cultural Heritage building materials. Specifically, a surfactant-assisted sol-gel synthesis to produce, in-situ on the building, crack-free nanomaterials to be used as long-term consolidants. Additionally, hydrophobic, water-repellent, self-cleaning, and biocidal properties can be incorporated into the product by innovative chemical modifications of the proposed synthesis route. Finally, I will summarize the future challenges of our group related to conservation of historic concrete in the framework of the Horizon 2020 project “InnovaConcrete”.

57 Towards the European Research Infrastructure in Heritage Science: E-RIHS L. Pezzati, CNR-Istituto Nazionale di Ottica The European Research Infrastructure for Heritage Science (E-RIHS) entered the European Strategic Roadmap for Research Infrastructures ESPRI Roadmap in 2016, as one of the six new projects. E-RIHS support research on heritage interpretation, preservation, documentation and management. Both cultural and natural heritage are addressed: collections, buildings, archaeological sites, digital and intangible heritage. E-RIHS is a distributed research infrastructure with a multi-level star-structure: facilities from many Countries will be organized in national networks, coordinated by separate National Hubs and RIHS headquarters will provide the unique access to all E-RIHS services, by coordinating the net of National Hubs.

58 Discussion & Conclusions

Modelling and monitoring of pesticides fate and exposure in a regulatory context (II)

59 Scenario Development for Off-field Soil Exposure and Risk Assessment M. Wang, WSC Scientific GmbH / Dept Efafe Modelling; J. Kleinmann, WSC Scientific GmbH; T. Schad, Bayer AG / Environmental Modelling; K. Hammel, Bayer AG, Crop Science Division / Environmental Safety; G. Ernst, Bayer AG / Ecotoxicology; G. Goenitz, Bayer CropScience AG / Environmental Safety; P. Neumann, Buyer Ag; S. Bub, Tiers Solutions GmbH

In its Scientific Opinion on risk assessment for in-soil organisms EFSA proposes a preliminary approach for off-field soil exposure by adding up entries from the different major exposure routes. EFSA indicates the worst-case character of its scenario “In the absence of appropriate off-field exposure scenarios…” and, hence, emphasises the necessity for model and scenario development. The present work aims to undertake first steps (i) to develop a model approach for off-field/crop soil exposure due to runoff, erosion, and drift, (ii) to develop exemplary schematic and real-world scenarios, (iii) which allow to gain insights in off-field soil exposure and risk using case studies. The developments are based on the Specific Protection Goals options. A tiered modelling approach is presented which allows to build exposure scenarios ranging from simple schematic and conservative to more realistic landscape-scale tiers, which can be easily linked to effect modelling (toxicological, population, community). Results are intended to support the design of off-field soil exposure and risk characterisation scenarios and the development of assessment endpoints relevant to specific GPS.

60 Biogenic residues formation from pesticides - an overview K. Nowak, TU Berlin / Institute for Environmental Research (Biology V); A. Miltner, Helmholtz Centre for Environmental Research UFZ / Department of Environmental Biotechnology; M. Kastner, Helmholtz centre for environmental health, WSC / Department of Environmental Toxicology; J. Kleinmann, WSC Scientific GmbH / Dept Efafe Modelling

Anthropogenic organic chemicals are deliberately (e.g. pesticides) released in major amounts to nearly all compartments of the environment. Soils and sediments as complex matrices provide a wide variety of binding sites and are the major sinks for these compounds. Pesticides entering these complex systems may undergo various turnover processes. They can be degraded chemically (e.g. photolysis), biodegraded by microorganisms, or volatilised leached to the groundwater or taken up by living organisms or immobilised in the form of non-extractable residues (NER). Microorganisms can use C and N from a pesticide to synthesize their biomass compounds, e.g. amino acids (AA) and fatty acids (FA). The extraction of known microbial biomarkers from soil or sediment after addition of C and N isotope tracer allows an estimation of microbial activity in the transformation of pesticide. We investigated the turnover of 2 pesticides (2,4-D, glyphosate, metamitron, bentazon, bromoxynil and clodinafo-proparglyl) with the particular focus on the metabolic incorporation of the isotope label into AA, FA and their fate over time. An agricultural soil and water-sediment were incubated with stable isotope labelled respective herbicide in the dark and at constant temperature (20°C). Soil and sediment samples at the respective sampling date were analysed for the amount and isotopic composition of AA, FA, CO2, solvent-extractable parent compound and metabolites and total NER. The presented data indicated that easily biodegradable herbicides e.g. glyphosate, 2,4-D or metamitron were utilized as a carbon (and
FOCUS scenarios were simulated and compared (five drainage scenarios with procedures. The modelling approach leveraged the aquatic environments. To make observed chemical concentrations for a wide range of species, chemicals, and whether a hydrophobic insecticide with log $K_{ow}$ is available, a default value is to be applied. An increase of this default value from 0.5 cm$^{-1}$ to 1 cm$^{-1}$ has been recommended by EFSAs, which results in more exhaustive wash-off from the plant surface. Generally, the extent of rainfall-induced displaced substance depends on several factors. An EC network group recommended a harmonized experimental approach to derive wash-off factors in the greenhouse: a 24th time interval between pesticide spraying and 10 to 20 mm of artificial rain, followed by an extraction of the plant material with an acetonitrile/water mixture of 80:20 (v/v). This study was conducted in order to derive a reliable numerical wash-off factor as input for FOCUS modelling. In this study, the published experimental data was reviewed for the availability of data suited for the calculation of a wash-off factor, reflecting a variety of different investigation types in terms of (time of (artificial) rainfall after application, rainfall amount and intensity, formulation, crops under investigation, etc. Published experimental wash-off studies are usually not conducted according to the standardized experimental procedures. Thus, only a limited number of the published studies are suitable to derive a wash-off factor for modelling. The outcome of the literature review presented herein suggests that a meaningful default wash-off factor should be well below 1 cm$^{-1}$. Keeping the existing default value of 0.5 cm$^{-1}$ retains a sufficient protection level while at the same time avoids a large number of unnecessary refinement studies.

Application of a dynamic aquatic food web model for FOCUS exposure assessment

Regulatory setting, the potential for bioaccumulation and biomagnification of plant protection product active substances in aquatic organisms is evaluated with simple screens on the basis of a substance’s $K_{ow}$, where typically a value greater than or equal to 3 indicates concern. However, this criterion may lead to false positive identification, because it does not account for biotransformation of the substance in biotic environment or bioavailability in the environment. Dynamic aquatic food web models are more refined tools for determining bioaccumulation and biomagnification potential, because they can account for chemical bioavailability and temporal and spatial variability in exposure concentrations due to seasonal and regional differences in weather and agricultural practices. The aim of this work is to demonstrate a modelling approach that couples standard FOCUS landscape and water body models with a dynamic aquatic food web model to assess whether a hydrophobic insecticide with $K_{ow}$ above the screening threshold of 3 will bioaccumulate/biomagnify. The Simon Fraser University (SFU) aquatic food web model, which predicts chemical concentrations in biota at six different trophic levels within an aquatic ecosystem, was selected based on the availability of data for relatively few input parameters and its demonstrated capability to predict observed chemical concentrations for a wide range of species, chemicals, and aquatic environments. To maximize relevance for agricultural systems in Europe, the food web model was adapted to accept environmental concentration time series input from the established TOXSWA model used in EU pesticide registration procedures. The modelling approach leveraged the transient form of the aquatic food web model and the time-varying parameters used to modulate the dynamic aquatic settings. Modelling results included both the daily time series predictions of organism concentrations for the six trophic levels and the uptake and elimination rate constants calculated from organism sub-models. In total, nine FOCUS scenarios were simulated and compared (five drainage scenarios with MACRO and four runoff scenarios with PRZM) and dominant organism uptake pathways were identified. The approach may be used to refine log $K_{ow}$-based screening bioaccumulation and biomagnification evaluations for regulatory purposes.

Improved assessment of pesticide peak exposure in cultivated mountain watersheds

The use of such chemicals can occur near surface water bodies, thus creating a potential for adverse effects on aquatic ecosystems. Due to the spatio-temporal variability of chemical applications and of the processes regulating their fate and transport to surface waters, ecosystems are often exposed to pulses of contaminants. In certain environmental scenarios, such as small mountain watersheds, where runoff fluxes are particularly rapid due to side slopes, exposure peaks can be shorter but much higher. Monitoring campaigns are often inadequate or too expensive to be carried out and modelling tools are therefore vital for exposure assessment and their use is encouraged by current legislation. However, currently adopted models and scenarios (e.g., FOCUS for PPPs) are often too conservative and/or “static” to accurately capture exposure variability, and the need for more realistic and dynamic tools is now one of the major challenges for risk assessment. In a previous work, the new fate model DynAPlus was developed to improve pesticide fate predictions in cultivated mountain basins. The model was successfully evaluated against chloropyrifos water concentrations measured in the Novella River (Valley, Northern Italy), where more than 1000 ha of apple orchards surround the river and Institute of Bioeconomy Research; P. Horney, Julius Kühn Institute; D. Daehmlow, Institute of Bioeconomy Research (NIBIO); E. Solbakken, R. Lågbu, Norwegian Institute / Institute for Strategies and Technology Assessment; B. Golja, Julius Kühn Institute / Institute for Strategies and Technology Assessment; J. Strassemeyer, ral Research Centre for Cultivated Plants / Institute for Strategies and Technology Assessment

In response to the implementation of the EU directive on sustainable pesticide use by Norway, the project SMARTCROP (funded by the Research Council of Norway), was started to address the challenges of developing and providing farmers with the necessary IPM tools. Towards this objective, SYNOPS, a risk indicator developed by the Julius-Kühn Institut, Germany, was adapted for Norway and provided with a graphical user interface such that a farmer or non-expert could perform risk assessments for field-specific pesticide applications. US-EPA PRZM5 and VFSMOM have been incorporated in SYNOPS for a more realistic modelling of the runoff/erosion modules and the functioning of the vegetated filter strip. The Norwegian tool, SYNOPS-Web, is available in both English and Norwegian. It uses Norwegian land-use, surface water and soil data, plant protection products registered for use in Norway, modified crop data for Norwegian conditions, and station-based weather data. Data can be provided for specific field and application scenarios. Another important new feature is the implementation of various mitigation measures such as vegetated filter strips, hedges, tillage/mulch, and cover crops. Risk assessments can be performed for a combination of mitigation measures in order to select the optimal application strategy under specific field conditions. In this presentation, we describe and discuss the mitigation measures implemented in SYNOPS-Web, Norway, and the corresponding adjustments to the model input parameters. We provide example scenarios based on realistic application patterns, without and with mitigation measures. Aquatic and terrestrial risk indices are presented to the user as Exposure Toxicity Ratios (ETR) in the form of colour-coded tables for an easy visual appraisal of the environmental risk under different conditions. In addition the predicted environmental concentrations can be viewed on a daily basis for the selected time period.
on their Environmental Fate and Effects (II)

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Acute Toxicity of Pyrene Associated with Dissolved Organic Matter of Various Molecular Weights to Daphnia magna

H. Liu, X. Xia, S. Bi, X. Jiang, H. Wang, W. Wen, School of Environment, Beijing Normal University

Dissolved organic matter (DOM) is a key environmental factor for the toxicity of hydrophobic organic compounds (HOCs) in natural waters. However, the toxicity of DOM-associated HOCs is still not clear. In this research, pyrene was selected as a model HOC and its toxicity as dissolved compounds (C_{Diss}) was measured by passive dosing systems. The immobilization and enzymatic activities of Daphnia magna were examined to analyze the toxicity of DOM-associated pyrene. The results indicated that the immobilization of Daphnia magna in the systems containing various molecular weight DOM and pyrene was ordered as middle molecular weight (MMW, 5-10k Da) DOM > higher molecular weight (HMW, > 100k Da) DOM > MMW < 1k Da DOM > 3-5k Da DOM. Furthermore, the superoxide dismutase (SOD), catalase (CAT), and peroxidase (POD) activity of Daphnia magna decreased gradually with the increasing C_{Diss} in the systems of MMW and HMW DOM, whereas increased when C_{Diss} was at a low level and then decreased when C_{Diss} was at a higher level in the control group with pyrene only and the system of LMW DOM. The influencing factors of Daphnia magna were related with the amount of pyrene sorbed on DOM, the uptake routes of DOM by Daphnia magna, and the desorption of pyrene from DOM in the gut of Daphnia magna. The findings obtained in this research suggest that the toxicity of DOM-associated HOCs should be taken into account for the eco-environmental risk assessment of HOCs in water systems.

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Passive dosing for constant concentration and defined composition of hydrophobic organic mixtures

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There is regulatory and scientific attention on the fate, exposure and effects of chemical mixtures including complex mixtures of hydrophobic chemicals such as petroleum substances and essential oils. These mixtures have recently been categorized as multicomponent substances and substances of unknown or variable composition, complex reaction products or biological materials (UVCBs). The dosing of such mixtures in environmental experiments and tests can be challenging and there is an urgent need for new methods to provide stable concentrations and defined composition of these mixtures in aquatic fate and toxicity testing. Passive dosing from silicone rods has successfully been used in biodegradation and toxicity testing of hydrophobic chemicals covering a broad chemical space in terms of K_{ow} and K_{oc}. This study aims to extend the applicability of the novel passive dosing method to hydrophobic multicomponent substances and UVCBs (i.e. complex mixtures). The method is straightforward: a silicone rod is loaded by direct addition of the mixture and subsequently equilibrated with ultrapure water to create constant and defined concentrations of each mixture constituent and thus also a constant and defined mixture composition. The aqueous concentration level can be controlled by the amount of mixture added to the rod. Early results show a good performance of the method with very fast dosing kinetics, aqueous concentrations increasing linearly with loading level and good reproducibility of the passive dosing for a petroleum substance and an essential oil. The presentation will focus on (1) the fast and reproducible loading of selected UVCB mixtures, (2) reproducibility and optimized passive dosing kinetics for one petroleum substance and one essential oil and (3) the performance and characteristics of the passive dosing method compared with more traditional dosing methods.

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Biodegradation of volatile substances in soil - Challenges and optimization of test setups (OECD 307)


Higher tier biodegradation laboratory tests in soil, sediment and/or surface water systems are conducted using standard OECD guidelines. As stated in these guidelines, they are not suitable for testing volatile chemicals, however a threshold based on Henry's law is not defined, except in OECD 309. In the actual setups, incomplete mass balance is a major problem while testing volatile chemicals. Optionally, OECD 307 and 308 allow biometer-type incubation setups but it does not require any data to prove if the systems remains aerobic. In addition, the degradation kinetics in a closed test system can largely be influenced by air-water partitioning as described by Birch et al. 2017. Our objective was to design a closed incubation test set up where maintaining and measuring of aerobic conditions was possible without the loss of test chemical. Additionally, a full scale OECD 307 with two model chemicals was performed to check the reproducibility of data in terms of mass balance and to better understand the obtained degradation data. The test setup consisted of 100 ml flask with 50g soil, CO2-trap and a Tenax tube completely closed using a stainless steel lock system. Oxygen saturation in the headspace was measured in a reference sample using optical measurements without the need to open the vessel. If the oxygen saturation was < 15%, the samples was aerated with oxygen-rich air. Applying this setup, degradation of 14C-labelled Tetrafluor and Decane was conducted on model soils with different texture, organic carbon and microbial activity. At sampling dates the headspace air of the samples was stripped off through the Tenax tube using a vacuum pump. The Tenax tube and the CO2-trap were taken for analysis and the soil was taken for extraction using appropriate methods. The solid extraction residue was subject to combustion analysis to determine the non-extractable residues (NER). The average overall recovery of 99.25% (N=19) for Decane and 104.34% (N=90) for Tetrafluor with a variation (between the individual replicates) < 11% for both studies suggests that obtaining a complete mass balance with the new test setup was reproducible. It was observed that radioactivity adsorbed on Tenax (100% parent), was higher in the soils with lower OC content suggesting that sorption of the chemical affecting its volatilization and hence degradation. Thus, how to deal with the volatilized parent fraction while calculating degradation kinetics is still a part of ongoing research.

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Untangling the biodegradation of hydrophobic chemicals in OECD and novel tests using a unified modelling approach

F. Polski, Technical University of Denmark (DTU) / DTU Environment; A. Brand, DTU Environment / DTU Environment; F. Sibany, K. Smith, RWTH Aachen University / Institute for Environmental Research; A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics; M. Kästner, Helmholtz centre for environmental research - UFZ / Department of Environmental Biotechnology; S. Trapp, Technical University of Denmark / DTU Environment

Persistence assessment using standardized (e.g., OECD) tests is one of the main challenges for hydrophobic organic chemicals (HOCs). While the use of radiolabeled chemicals brought major advances, mechanistic models can still provide deeper insights in experimental results and underlying processes. In this context, the key objective of this study was to test the applicability of a unified modelling approach across the spectrum of OECD degradation tests and newly developed experimental tests for HOCs. We specifically aimed at (i) elucidating biodegradation kinetics and improving their estimation by including a new method for microbial yield calculation; (ii) determining 14C fractions (mineralized, incorporated in biomass and non-extractable residue NER) at the end of tests as persistence indicators. The unified model for sorption and biodegradation (in combination with the Microbial Turnover to Biomass growth yield estimation method) was used to predict mineralization to CO2 growth of degrading microorganisms and NER formation in aerobic degradation tests with selected 14C-labeled HOCs (triclosan, pentachlorophenol—PCP, propargite and pyriproxyfen). Model predictions were fit to experimental results obtained elsewhere in conventional degradation tests activated sludge or soil in novel passive dosing setups, in which we aimed for a tight agreement between model predictions and empirical data was shown by adjusting only the ratio v_{break}/K_w, which describes biodegradation kinetics according to the Michaelis Menten equation. Overall, a high range of v_{break}/K_w values was shown for the selected substances (0—55 mg l^{-1}d^{-1}), indicating that both limited bioavailability and intrinsic recalcitrance can explain HOC persistence. This study represents a first attempt of using a unified modelling approach for predicting biodegradation of HOCs across a variety of tests, showing promising results towards persistence prediction of organic chemicals during regulatory screening. Ongoing research is focusing on extending the model applicability by (i) including the formation of intermediate transformation products; (ii) determining (de)sorption limitation based on dedicated experiments; and (iii) using uncertainty-based approaches to support decision makers within REACH.

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History of polychlorinated biphenyl deposition to snow and ice from the Lomonosovfonna glacier, Svalbard

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Polychlorinated biphenyl (PCB) deposition to the north polar regions has long been recognized. Here we investigate quantitative and qualitative PCB inputs to the Lomonosovfonna glacier on Svalbard from an ice core drilled in 2009, and a snow pit from spring 2010. Lomonosovfonna is the highest ice on Svalbard at 1250 masl. It is above the tropospheric boundary layer at all times of year and there is an urgent need for new methods to provide stable concentrations and defined composition of DOM. Furthermore, the superoxide dismutase (SOD), catalase (CAT), and peroxidase (POD) activity of Daphnia magna decreased gradually with the increasing C_{Diss} in the systems of MMW and HMW DOM, whereas increased when C_{Diss} was at a low level and then decreased when C_{Diss} was at a higher level in the control group with pyrene only and the system of LMW DOM. The influencing factors of Daphnia magna were related with the amount of pyrene sorbed on DOM, the uptake routes of DOM by Daphnia magna, and the desorption of pyrene from DOM in the gut of Daphnia magna. The findings obtained in this research suggest that the toxicity of DOM-associated HOCs should be taken into account for the eco-environmental risk assessment of HOCs in water systems.
Environmental occurrence and distribution of organic UV stabilizers in the sediment of the Bohai and Yellow Seas

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Organic UV stabilizers are of emerging environmental concern due to their large production volumes and potential endocrine disrupting properties. UV stabilizers are widely used in plastic products, paints and coatings to improve the product stability against UV light. Furthermore, some UV stabilizers are approved as ingredients in personal care products like cosmetics and sunscreens. The pathways into the marine environment are either indirect by wastewater treatment plant discharges or direct by recreational activities like bathing and swimming. Four benzotriazole UV stabilizers are classified as SVHC (Substances of Very High Concern) under the EU legislation REACH. Numerous others are currently listed under the European community rolling action plan (CoRAP) to be re-evaluated in the next years. Due to their chemical properties, most UV stabilizers accumulate in sediment (logKow > 3) and have potential for persistence or pseudo-persistence.

Environmental data for the coastal and marine environment are sparse. For this study 74 surface sediment samples of the Bohai and Yellow Seas were analysed for 19 commonly used organic UV stabilizers. The sample pretreatment and analysis was carried out as follows: First, the samples were homogenized with sodium sulphate. Afterwards, extraction and clean-up was performed using an accelerated solvent extraction (ASE-350, DIONEX, Germany) method. For this, 22 mL stainless steel ASE cells were filled with 3 g of 10% deactivated silica and approximately 5 g sediment that was spiked with appropriate isotopically labelled standards. The cells were extracted using dichloromethane for three 15 min-cycles at 100 °C. The extracts were solvent-cleaned to methanol, reduced in volume to 150 µL. The instrumental analysis was performed on a LC-MS/MS system (1290 Infinity coupled to 6490 triple quadrupole LC/MS, Agilent Technologies, Germany) equipped with an APPI-source and a C18 column (Eclipse Plus RRHD 1.8 µm, 2.1 x 150 mm, Agilent Technologies, Germany). This study shows levels of contamination and distribution of organic UV stabilizers in surface sediments of the Bohai and Yellow Seas for the first time. 14 substances have been positively detected in concentrations in the low ng/g dw range. Characteristic pollution profiles and distribution pattern have been identified, which indicate to different indirect sources of UV stabilizers into the study area.

LCIA method developments in a global perspective: Status and outlook (I)

71 Implications of spatial differentiation on LCA-based decision-making: a case study of biochar systems in Indonesia

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The development of spatially differentiated life cycle impact assessment (LCIA) methods and their use in regionalized life cycle assessment (LCA) has intensified in the past few years. However, it is less investigated whether spatial differentiation leads to more correct decisions based on the LCA, in addition to just more accurate and realistic LCIA results. The main goal of this work was to therefore assess the implications of spatial differentiation on the interpretation phase of a comparative LCA. Biochar production from biomass residues and its use as soil conditioner in Indonesia was used as case study. Comparisons were made between 4 villages, 3 biochar production techniques, and 2 fertilization strategies. Results showed that (i) regionalized impact scores for individual impact categories either increased or decreased compared with site-generic scores, depending on the impact category (by up to 1 order of magnitude); (ii) total damages to human health were approximately 3 to 5 times higher when compared to site-generic scores and (iv) irrespective of the geographic locations, regionalization of damages contributed or detracted to site-specific scores. This is mainly because of trade-offs between categories, where increase in impact scores for some categories was compensated by decrease in others. Overall, irrespective of the approach to spatial differentiation in LCIA, biochar production and use in agriculture is generally expected to bring environmental benefits. When parameter and inventory uncertainties were considered, there was no indication of substantial differences in the potential of best performing villages in terms of total damage to human health and ecosystems, although village performing worst with regard to total damage to human health changed. There was a general tendency that biochar production using both Kon Tiki and Adam retort kilns performed better than earth-mound kiln, and furthermore biochars brought largest benefits where no-biochar agricultural production systems were based on inorganic fertilizers. This rather consistent ranking was mainly due to relatively large geographic differences in life cycle inventories between villages, which were often larger than geographic differences in characterization factors between site-specific and site-generic approaches. Thus, although spatial differentiation improved accuracy and realism of environmental impacts in this comparative case study, it did not necessarily contribute to more correct decisions.

72 Considering space debris related impacts within the LCIA framework

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The space sector is a new area of development for LCA studies. The European Space Agency (ESA) has been working since 2012 on environmental issues for space activities through its Clean Space Initiative. ArianeGroup, which is the prime contractor for the development of the new Ariane 6, is currently performing an LCA of this launcher in exploitation phase (ESA’s contractual requirement). However, the current studies adopt a Cradle-to-Launch pad approach due to lack of relevant modelling for use and disposal phase. In addition, a rising sustainability concern is occurring in the space sector particularly regarding impact of space debris: 29,000 human-made objects, larger than 10cm, are orbiting the Earth but only 6% are operational spacecraft, being today a significant and constant danger for all space missions. Consequently, considering end-of-life management during the design of spacecraft becomes a subject of crucial importance. Given this situation, there is an opportunity to make the link between space debris concern and eco-design of spacecraft (satellites & launchers) using the LCA methodology. A focus should be put on the comparison of several missions & post-mission disposal scenarios to study potential trade-offs between typical impact categories (e.g. toxicity and climate change), but also with regard to the growing issue of space debris. Hence, our challenge is to integrate, via a dedicated additional indicator, space debris related impacts within the LCIA to broaden the scope of LCA for space systems. The Area-of-Protection Resources has been identified to reflect the depletion of available orbits by the potential generation of space debris. Considering generation of debris in operating orbits with a resource depletion perspective allows us to address the environmental footprint of the spacecraft’s design. Volume occupation by debris and dead spacecraft leads to a decrease of the orbital resource availability enhancing the risk of collision/break-up and then propagation of new clouds of debris. As a consequence, the lack of access to the orbital resource in the future (scarcity) could be handled as environmental and socio-economic impacts. The presentation will prove the relevancy of this approach by presenting the impact pathway linking the occupation of operational orbits and environmental impacts. Environmental mechanisms and impacts (midpoints) will be exposed with associated specific characterization factors.

73 Implementing ozone formation effects due to poplar plantations for biomass production in Europe as an impact assessment metric

P. Vercoulen, Radboud University; R. Kranenburg, C. Hendriks, TNO; R. Van Zelm, Radboud University / Department of Environmental Science

Poplar trees are known to emit volatile organic compounds, among them isoprene, which has been positively linked to ozone formation. Given this situation, there is an opportunity to make the link between space debris concern and eco-design of spacecraft (satellites & launchers) using the LCA methodology. A focus should be put on the comparison of several missions & post-mission disposal scenarios to study potential trade-offs between typical impact categories (e.g. toxicity and climate change), but also with regard to the growing issue of space debris. Hence, our challenge is to integrate, via a dedicated additional indicator, space debris related impacts within the LCIA to broaden the scope of LCA for space systems. The Area-of-Protection Resources has been identified to reflect the depletion of available orbits by the potential generation of space debris. Considering generation of debris in operating orbits with a resource depletion perspective allows us to address the environmental footprint of the spacecraft’s design. Volume occupation by debris and dead spacecraft leads to a decrease of the orbital resource availability enhancing the risk of collision/break-up and then propagation of new clouds of debris. As a consequence, the lack of access to the orbital resource in the future (scarcity) could be handled as environmental and socio-economic impacts. The presentation will prove the relevancy of this approach by presenting the impact pathway linking the occupation of operational orbits and environmental impacts. Environmental mechanisms and impacts (midpoints) will be exposed with associated specific characterization factors.
LOTOS-EURAS on a 0.5x0.25° scale. The effect factor was based on a lognormal relationship between the PAF of plant and grassland species and ground-level AOT40. To test our developed CFs, a case study was performed of electricity generation in a coal power plant for varying fuel mixtures of hard coal and poplar biomass. The functional unit was 1 kWh of electricity generated. ReCiPe2016 was used for the impact assessment of other impact categories. Largest CFs were shown in southern and central parts of Europe, which is mainly caused by the effect factor of these two regions being 0.1. For electricity we found 7.6·10⁻³ km² eq kg⁻¹ yr⁻¹ km² eq urb⁻¹. The area-weighted European average CF was 2.9·10⁻³ km² eq yr⁻¹ km² eq urb⁻¹. Case study results show that ozone formation caused by cultivation of poplar plantations can contribute significantly to overall ecosystem impacts due to electricity generation from poplar biomass. When fully using biomass for electricity production, cultivation of poplar plantations in Italy even contributed 20% to the total adverse effects to ecosystems. With our work ozone formation impacts due to poplar tree plantations for biomass production can be quantified. We showed that contributions to ecosystem damage from these emissions in an electricity generation case study were not negligible. Therefore, we recommend to include the CFs in future LCA case-studies where poplar plantations are included.

74 Relative potency approach for using in vitro information for calculating human effect factors in LCA
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Although manufactured nanomaterials (MMNs) offer several advantages compared to their bulk form, several concerns have been raised on their environmental and human risk. The LCA methodology is a valid tool to assess the environmental and human impact of nanoproducts. But overall, LCA studies of MMNs and nanotechnology are currently affected by a gap of knowledge regarding the exposure and toxicity of MMN releases into the environment during different life cycle stages. Within the LCA methodology, the human toxic effect (EF) is evaluated based on ED₅₀ extrapolated by in vivostudies or human studies. In vision of the “Toxicity Testing in 21st Century”, the in vitro tests are expected to be replaced by in vitro studies. Also, for emerging materials such as nanomaterials, still a scarce number of in vivo data are available in literature. Given the expansion of in vitro testing, there is probably a potential to use outputs from such in vitro testing in order to derive ED₅₀ values for the use in LCA studies. Here, we propose to integrate in vitro data in the assessment of the human toxic potential using a relative potency (RP) approach. The RP approach has been widely used by toxicologist for ranking chemical, to calculate the equivalent dose of one chemical that produce the same response as another at a specific dose or to define toxic equivalency factor. In vitro studies for one and the same response have been performed on 2 cells and HepG2 cell line. Based on the EC₅₀ values obtained by the in vitro test a RP factor has been calculated and used to calculate the ED₅₀ for the above mentioned nanomaterial. Therefore, we present a first attempt for the derivation of EF values for their implementation into LCIA factors for human toxicity, by using in vitro data values for nano-CoO, nano-Ag and nano-ZnO in combination with a relative potency (RP) approach. Until more comprehensive toxicity data (i.e. ED₅₀) as well as a more sophisticated method to convert in vitroto in vivo data become available, we can consider in this study applied procedure as the good approximation in order to make use of the already large and continuously increasing body of in vitro toxicological studies on nanoparticles and like this allow their use in the field of LCA.

75 Integrating endocrine disruption into life cycle impact assessment
Y.A. Emara, A. Lehmann, M. Siegert, Technische Universität Berlin / Chair of Sustainable Engineering; M. Finkbeiner, Technische Universität Berlin / Chair of Sustainable Engineering
Converging lines of evidence suggest that exposure to ‘endocrine-disrupting chemicals (EDCs)’, i.e. chemicals with the ability to interfere with and alter functions of the endocrine system, is linked to multiple adverse effects on humans and wildlife (e.g. diabetes and reproductive dysfunctions). Currently, life cycle impact assessment (LCIA) models targeted at characterizing toxicity-related impacts of chemicals do not model EDC toxicity. This study proposes a new approach to include endocrine disruption (ED) as a new impact pathway within LCIA and establish two new impact categories (Human ED and wildlife ED), thereby capturing adverse endocrine-mediated effects on humans and ecosystems separate from other toxicity-related impacts. Relying on the USEtox model, the calculation of fate and exposure factors remains unchanged, while the effect factor is determined using effect data on several ED-sensitive toxicological endpoints, thus reflecting the spectrum of endocrine mechanisms by which an EDC is known to act (e.g. estrogen receptor antagonism or interference with thyroid pathways) and the resulting mosaic toxic effects. To overcome potential data constraints in finding suitable toxicological effect data for the thousands of chemicals suspected to exhibit endocrine-disrupting characteristics, data provided by the United States Environmental Protection Agency on the basis of in-vitro high-throughput screening assays for the endocrine bioactivity of more than 1,000 chemicals is examined for its suitability to be used in the proposed approach. New characterization factors for a set of known EDCs will be provided to directly enable characterization of EDCs and their adverse effects within future life cycle assessment studies.

76 Poster spotlight: MO090, MO091, MO100

Environmental fate of emerging contaminants in the water cycle: analytical challenges and engineered solutions (II)

77 Review on removal and reactions of micropollutants in biofilms under growth and non-growth conditions
K. Bester, Aarhus University / Environmental Science; M. Escolà Casas, Aarhus University / Department of Environmental Sciences; U. Bollmann, Aarhus University / Environmental Science; P. Carvalho, Aarhus University, Department of Environmental Science / Department of Environmental Science; E. Torresi, Kruger A/S; H. El-taliawy, Aarhus University / Department of Environmental Science; L. Zhang, Aarhus University / Department of Bioscience; G. Ooi, K. Tang, DTU Environment; H.R. Andersen, Technical University of Denmark / Department of Environmental Engineering; M. Christenson, Anox Kaldnes Pharmaceuticals and other compounds need to be removed from wastewater. This contribution will give an overview on the possibilities of removing micropollutants with biofilms. Biofilms occur in nature, but are also increasingly used in technical installations to remove micropollutants from water (wastewater and drinking water), porous media biofilm systems (sandfilters), moving bed biofilm systems (MBBRs). Biofilms can remove considerably better than suspended bacteria: diclofenac is recalcitrant in sludge systems, while it can be degraded with half-lives of 2 h in biofilm systems. In this contribution it is highlighted which ecological conditions (aerated versus denitrifying; high and low BOD loads) have been found to favor degradation. It was found that BOD supply has massive impact on the removal: On the one hand high BOD loads led to enhanced growth of biofilm, resulting in high turnover and reaction rates also of the respective micropollutants. On the other hand the biomass related reaction rate constants are considerably higher with low BOD loads. The highest reaction rate constants and biomass related reaction rate constants were found for systems with intermittent BOD loading. The switch from aerated to denitrifying conditions, however only gives effects for a chosen few compounds like ibuprofen, while reaction rates usually are similar. So the redox conditions as such are obviously not critical. Biofilms have often proven to have unique degradation pathways leading to hitherto strange metabolites. On the other hand it is not always the case that biofilms are capable to reach high removal rates for otherwise persistent ozonation-by-products such as macrolide N oxides while avoiding back reactions or incomplete biodegradation. This also holds for most of the ozonation products of diclofenac.

78 Biodegradation of emerging organic contaminants using an enzyme-mediator system and study of the resulting transformation products
I. Caraena, Curtin University / chemical department; C. Joll, Curtin University / chemical department; K. Linge, Y. Gruchlik, Curtin University; A. Paparini, Murdoch University
Due to improvements in analytical screening methods, a large number of emerging organic contaminants (pharmaceutically active compounds, personal care products, pesticides, surfactants, plasticizers, corrosion inhibitors, flame retardants, artificial sweeteners and others) have been identified in the aquatic environment. Biodegradation is one of the processes that can remove potentially hazardous emerging organic contaminants from different environments, with the help of microorganisms (e.g. algae, bacteria or fungi) and their extracellular products, in both aerobic and anaerobic conditions. The objective of this study was to investigate the biodegradation of a series of antibiotics and one anticonvulsant using laccase enzyme, extracted from a white-rot fungi Trametes Versicolor, in the presence of 2,5-diamino-3(ethylbenzothiazoline-6-sulfonic acid) diammonium salt (ABTS), as a mediator. The experiments were conducted in MilliQ water at different pH values, with different ratios of laccase and mediator, under aerobic and static conditions. The degradation was monitored by measuring the concentration of the remaining antibiotic over 168 hours, using a high performance liquid chromatograph with UV detection. The formation of new peaks was also monitored.
Dispersed into the microorganisms in sewage and successfully removed by constructed wetland systems (CWs). The aim of the present study was to investigate the transformation of antibiotics with high resolution mass spectrometry. The validated method was proven to be reliable, thanks to the combination of two quantification approaches, i.e., matrix solid-phase microextraction (SPME) fiber coupled to liquid chromatography-tandem mass spectrometry (LC-MS) as well as to stable isotope dilution analysis (SIA-MS). The three antibiotics studied were characterized using Fourier transform infrared (FTIR) spectroscopy, scanning electron microscopy (SEM), and energy-dispersive X-ray (EDX) analysis in order to understand the removal mechanisms of the tested contaminants. The results showed that 89% degradation of sulfamethoxazole can be achieved at pH 5, with an enzyme activity ranging between 110-120 U/L and an initial mediator concentration of 200 µM, while trimethoprim only degraded by 43% under the same conditions. The results of similar degradation experiments on other antibiotics will be discussed in this conference presentation. The identification of transformation products of the antibiotics with high resolution mass spectrometry, and the microbial activity of the transformation products, will also be presented. This study provides a better understanding of the biodegradation of emerging organic contaminants and their transformation products. Further work can be done to assess the possible health and environmental risks associated with the reuse of treated wastewater, for applications such as irrigation and groundwater replenishment.
plant tissue was documented. Formation of transformation products was assessed, but the mass balances were not closed. Organic micropollutants sorption to support matrix was low. Removal of different compounds was higher in summer than in the winter. Planted reactors showed higher efficiency than unplanted reactors, stressing the synergies between the plant and the microbial community. Unsaturated systems tended to be more efficient. Removal correlated with the nitrification activity and with the biofilm activity, suggesting that ERDA as play an active role in the micropollutants biodegradation. The removal of the organic micropollutants in CWs is affected by several design and operational parameters. Plant uptake does occur but phytoaccumulation is low as the compounds can be degraded inside the plant tissues. Due to overlying effect of the plants, the extent of microbial degradation could not be quantified. Further studies on transformation products in this type of technical systems are needed.

Wildlife ecotoxicology; laboratory dosing studies to field population assessments (II)

83 Effects of PAH exposure on fuelling ability in a long distance migratory shorebird

K. Bianchini, University of Saskatchewan - Toxicology Centre / Toxicology: C.A. Morrissey, University of Saskatchewan / Biology

Many shorebirds are long distance migrants that stop to refuel along the journey where they can be exposed to pollutants that may impede fuelling for migration. Exposure to organic pollutants can cause potential effects on migration success, speed and subsequent population parameters since pre-migratory fuelling is correlated with reproductive performance upon reaching the northern breeding grounds. The polycyclic aromatic hydrocarbons (PAHs) found in marine oil pollution have the potential to interfere with pre-migratory fuelling physiology in shorebirds. However, a link between PAH exposure and pre-migratory fuelling has yet to be established. Our objective was to determine if PAHs or associated contaminants can affect condition and fuelling rates in a captive shorebird, the Sanderling and in the field at major shorebird stopovers. In this study, a captive population of 49 Sanderling (Calidris alba) was orally dosed with a commercial PAH mixture for 21 days at ecologically relevant concentrations (0, 12.6, 126, and 1260 μg/kg). We found that PAH exposure and pre-migratory fuelling were significantly elevated in the high dose group relative to controls and fuelling rates and condition were also lower in dosed birds. Higher PAH exposures were associated with reduced serum bile acid concentrations, elevated serum creatine kinase concentrations, and with high serum lipase concentrations (in females). These results suggest that PAH exposure can interfere with lipid transport and metabolism and can cause muscle damage leading to poorer condition. We also captured Sanderling from the Gulf of Mexico, which is subject to recurring oil spills and from Chaplin Lake, Saskatchewan, a relatively uncontaminated site. We measured each bird’s body condition, fuelling status, and plasma PAH levels and attached miniature radio transmitters to a subset of birds (n=75). Motus radio telemetry array technology was used to determine the arrival and departure timing and stopover duration. We found that stopover durations in the Gulf of Mexico were longer than in Chaplin Lake (27 versus 15 days), and that stopover duration was associated with body condition and fuelling status at capture. We also measured higher plasma PAH concentrations in birds at certain Gulf sites, suggesting that PAH exposure is associated with lower pre-migratory fuelling rates. This work will inform shorebird conservation by providing valuable insight into a potential cause of migratory shorebird declines.

84 PFAAs levels, oxidative status and reproductive success in great tits (Parus major) inhabiting a contamination hot-spot.

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Perfluorooalkyl acids (PFAAs) are substances which have been produced for more than five decades. Their unique properties of repelling both water and oil, make them suitable for many industrial and consumer applications such as water and dirt repellents for fabrics and carpets, active components in firefighting foams or precursors in Teflon® production [1]. Its extensive use, together with their high persistence, has resulted in global contamination of the environment, wildlife and even humans [2,3]. This ubiquity contrasts sharply with the limited amount of available data on their effects on non-target organisms. We report here the PFAAs egg and plasma levels in wild populations of great tits (Parus major) settled along an established pollution gradient starting from a fluorochromal plant in Antwerp (Belgium). Using two generations of great tits we have obtained important results in some poorly known issues such as the differences between sexes, maternal transfer of compounds or possible effects on the oxidative status or the reproductive success. The levels we detected in eggs and plasma, demonstrate that Antwerp is one of the major hot-spots in the world for perfluorinated compounds pollution. With regard to the possible effects, negative correlations were observed between PFAAs levels in the eggs and reproductive parameters, including the total hatching success, eggshell thickness or the total breeding success. PFAAs levels in blood correlated with protein damage in adult birds while in chicks they correlated with higher activity of antioxidant enzymes (GPX and CAT). The obtained data represent an important step towards the understanding of the behaviour, effects and consequences of PFAAs in wild bird populations. [1] Buck RC, Franklin J, Berger PE, Cameron P, Karper BM, S. Van Leeuwen SP (2011). Perfluorooalkyl and perfluoropolyalkyl substances in the environment: terminology, classification, and origins. Integ Environ Asses 7: 513-531. [2] Giesy JP and Kannan K (2001). Global distribution of perfluoroocante sulfonate in wildlife. Environ Sci Technol 35: 1339-1342. [3] Giesy JP and Kannan K (2002), Peer-reviewed: perfluorochemical surfactants in the environment. Environ Sci Technol 36: 146-152.

85 Active and passive monitoring of lead poisoning in birds of prey in Spain

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Lead is a toxic metal that is a great threat for birds of prey, presenting a relevant risk for their health and survival. The integration of active and passive monitoring permits to have a complete picture of the risk of Pb contamination for the different species. Here we present the results of the active monitoring performed between 2009 and 2016 with 14 unhatched eggs of West Canarian common kestrels from Tenerife Island collected between 2009 and 2016. We performed a passive monitoring by measuring blood (n=27) and liver (n=685) lead levels in birds of prey of 16 species found dead or sick in Spain between 2004 and 2017, but also an active monitoring by measuring blood lead levels in birds (n=196) exposed to lead pollution. We observed clear differences in blood Pb exposure between the specie with lower blood Pb levels (<200 μg/ml) and the specie with high blood Pb levels (>30 μg/ml). We also measured the Pb concentration in liver of the species with higher Pb levels in liver associated with clinical poisoning (18-30 μg/ml) d were cinereous vulture (1/3), Eurasian griffon vulture (87/118), Spanish imperial eagle (1/6) and red kite (1/18) presented elevated blood Pb exposure levels (>200 μg/ml). Passive monitoring revealed that the species with lead levels in liver associated with clinical poisoning (18-30 μg/ml) were cinereous vulture (1/3), Eurasian griffon vulture (2/228) and western marsh-harrier (1/3); and the species with clinical severe poisoning (>30 μg/ml of Pb in liver) were Eurasian griffon vulture (19/228), red kite (1/129) and golden eagle (3/36). The study of biomarkers reveals a negative relationship between Pb exposure in blood and liver Pb concentrations. Plasma lead concentrations were also affected by Pb exposure, because elevated blood Pb levels were associated with lower Pb levels in blood and higher Ca/P ratio in plasma of birds. Carotenoid levels in plasma were also increased in birds with higher blood Pb levels, indicating a possible allocation of antioxidants in plasma to cope with adverse effects of Pb. The integration of active and passive monitoring permits to have a complete picture of the risk of Pb contamination for the different species. Here we present the results of the active monitoring the elevated blood Pb levels (73.7% with >200 ng/ml) in field-trapped Eurasian griffons as found in previous studies, but also report a significant mortality (8.3% with >30 μg/ml d) in Eurasian griffins and golden eagles with the passive monitoring.

86 Persistence of elevated p,p'-DDE levels and HCB-related protoporphyrin IX decrease in eggs of common kestrels from Tenerife (Canary Islands, Spain)

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Persistent organochlorines (OC) pesticides, including p,p'-DDE, have been banned in many parts of the world for more than 30 years, but they are yet present in the top predators of terrestrial and aquatic food webs. The Canary Island were one of the Spanish regions with the highest use of OC pesticides due to the intensity of its agriculture. A previous study performed between 1988 and 1994 with 14 unhatched eggs of West Canarian common kestrels from Tenerife Island showed elevated concentrations of p,p'-DDE (17.9 μg/g dw; equivalent to 4.9 μg/g ww). Here, we have monitored the levels of OC compounds (pesticides and polychlorinated biphenyls) in 40 unhatched eggs of West Canarian common kestrels from Tenerife Island collected between 2009 and 2016. We have also monitored the levels of the carotenoid pigments in the eggs, which are used in different stages of life of the chick (eggs after hatching stage; Porphyrin difference and resuspension in n-hexane, followed by four clean-ups with sulfuric acid and determination by GC-ECD. For porphyrin determination, eggshells were homogenized and extracted with acetonitrile:CH3CN (2:1) and then
analysed by HPLC-DAD. Egg content showed the following OC levels (mean ± SE; µg/g dw): p,p'-DDE, 15.2 ± 1.7; p,p'-DDT, 0.118 ± 0.020; PCBs, 0.459 ± 0.121; HCHs (hexachlorocyclohexane isomers), 0.021 ± 0.003; and HCB (hexachlorobenzene), 0.004 ± 0.0004. p,p'-DDE levels have remained elevated for more than 20 years and these levels were statistically associated in general linear models with the surface of active and abandoned cropland in a 200 m-radius around the nest (+), distance from nest to urban areas and greenhouses (-, altitude (+) and year (highest in 2011). PCB levels were associated with distance from nest to roads (-) and altitude (+). The shell index was not affected by p,p'-DDE levels, but decreased with embryo development. Protoporphyrin IX was the only pigment in eggshells and its concentration was negatively affected by HCB levels in egg content.

87 Long-term increase in secondary exposure to anticoagulant rodenticides in European polecats in Britain K.A. Samsbury, University of Exeter / Environment and Sustainability Institute; R. Share, Centre for Ecology & Hydrology (NERC); H. Schofield, L. Crosse, The Vincent Wildlife Trust; M. Perera, Centre for Ecology & Hydrology / Lancaster; D. Sleep, NERC Centre for Ecology & Hydrology; A.C. Kitchener, G. Hankie, National Museums Scotland; R. McDonald, University of Exeter / Environment and Sustainability Institute

As a result of legal protection and population recovery in Great Britain, European polecats (Mustela putorius) are expanding into areas associated with greater usage of second-generation anticoagulant rodenticides (SGARs). We analysed livers from polecats found dead (mostly road casualties) from 2013-2016 for residues of five SGARs. We related variation in residues to polecat traits (age, sex, provenance), to potential exposure pathways by analysing stable isotopes of carbon (δ13C) and nitrogen (δ15N) in whiskers, and to data collected from polecats in the period 1992-99. In all, 54 of 68 (79%) polecats from 2013-16 had detectable liver residues of at least one SGAR. Bromadiolone (71%) was the most commonly detected compound, followed by difenacoum (53%) and brodifacoum (35%). Liver SGAR residues did not vary with sex or with the season in which the polecat died. We found a positive association between occurrence of liver SGAR residues and δ15N values. Polecats in Britain feed predominantly on rats and rabbits and our findings are consistent with the concept that individuals feeding on rats (higher trophic level than rabbits) are more likely to be exposed to SGARs. Total SGAR liver concentrations were higher in polecats from arable than pastoral habitats, consistent with more intensive SGAR use on arable farms, and higher in western than eastern regions although the reason for this is unclear. Both number of compounds and total SGAR concentrations were positively associated with age, presumably due to multiple sub-lethal exposures during an animal’s lifetime; older animals may thus be at most risk from poisoning due to progressive accumulation of liver residues. When we compared data for polecats from 2013-16 with those for polecats that died in 1992-99 and accounted for differences between studies in detection limits, we found that the rate of detection of SGARs in polecats in Britain increased 1.7 fold over the 25 year period. This increase was not restricted to newly recolonised areas and suggests an increase in the risk to polecats from SGARs throughout their range.

88 Poster spotlight: M0035, M0036, M0083

Environmental risk assessment in time- space - new approaches to deal with ecological complexity

89 The threshold option, the recovery option and landscape modelling P. Thorbek, Syngenta / Environmental Safety; N. Galic, Syngenta / Environmental Safety; V. Forbes, University of Minnesota / Ecology, Evolution & Behavior

Landscape provides a multitude of ecosystem services, but the relationships between the populations of the organisms providing them, stressors and the delivery of ecosystem services are rather complex and often nonlinear. In order to understand how pesticides may affect ecosystem services and biodiversity at the landscape scale, it is necessary to understand both exposure and effects at the organism level, but also how life history, movement patterns and farming activities such as tillage and harvest affect population dynamics. If this has to be done for all species in all landscapes in Europe the modelling task quickly becomes unmanageably complex, and the interpretation of the modelling outputs will be challenging. Here we present a tiered system for model design to aid managing the complexity. We outline what model design features are necessary for modelling based on species mobility and whether the ecological threshold option (ETO) or the ecological recovery option (ERO) is chosen. Ecological production functions quantitatively link the service-providing units to the services delivered and are therefore the basis of assessing the quality of population attributes (e.g. biomass or functioning) contribute to the final ecosystem services enjoyed by the recipients. Such understanding can be used to set the protection goals for different service-providing units for both ETO and ERO. The attributes which link to service delivery can be difficult to measure at the landscape level, but by combining ecological models and ecological production functions, thresholds can be set for lower tiers of the risk assessments, which may be easier to measure. In some cases the ecological production functions are quite simple if a population directly delivers the service (e.g. for angling). However, in other cases, the link is far from straightforward and such ecological production functions have largely been ignored in pesticide risk assessment. This should be a priority area for future research.

90 Understanding risk - a better approach to reduce uncertainty M. Wang, WSC Scientific GmbH / Dept Efate Modelling; M. Foudoulakis, Dow Agrosciences / RSRA ERS

For many compounds the intrinsic toxicity as determined in toxicity studies does not reflect toxicity and risk adequately. Rather other mechanisms determine which species are most at risk (local species) and how large the risk posed to these species is. These include for example elimination rates and feeding behaviour, which are not considered in the first tier. In the present presentation results from two case-studies are given which demonstrate how uncertainty in the risk assessment can be reduced by trying to understand mechanisms that lead to toxicity and mechanism determining the actual and long-term risk of mammals and birds in the field. Field data help to verify the obtained knowledge and to determine an empirical margin of safety. Finally, population modelling is used to answer what-if questions and to answer questions on the relevance of effects when considering specific worst-case assumptions. In both example compounds metabolism and excretion together with feeding behaviour mainly determined the acute and long-term risk. All of these mechanisms are not considered in the first tier risk assessment and without these it would not be possible to understand the risk of the compounds shown here. This understanding significantly reduced the uncertainty of the risk assessment, because with the gained knowledge it is possible to identify critical scenarios.

91 Developing spatio-temporally realistic representations of agricultural landscapes for assessing the impacts of pesticides on non-target organisms E. Ziółkowska, Jagiellonian University / Institute of Environmental Sciences; C.J. Topping, Aarhus University / Department of Bioscience; A. Bednarska, Polish Academy of Sciences / Institute of Nature Conservation; R. Laskowski, Jagiellonian University / Ecotoxicology & Stress Ecology Group

Species richness and population sizes in agro-ecosystems have decreased dramatically during the last decades. The current scenario of increasing agricultural intensification resulting in landscape simplification is considered one of the main causes of this biodiversity loss, along with widespread use of pesticides. As the management of landscape heterogeneity seems to be crucial for maintaining vital populations in agro-systems, it is necessary to include the landscape component in ERA and as the important mitigation strategy. We present a methodological framework for modelling the spatio-temporal heterogeneity in agricultural landscapes. The framework has been implemented within the ALMaSS simulation system allowing to investigate the effects of changes in landscape structure and management on the population size and distribution of animals. We describe spatial landscape heterogeneity through a detailed land cover map, in which farmed areas are represented as accurate maps of fields grouped into farm units of different types (e.g., pasture, arable farms). The assessment of the heterogeneity refers to both crop management throughout a year, described through individually tailored management plans for each crop, and the cropping system understood as a pluriannual crop rotation. Crop management plans consist of combinations of farm activities (including pesticide treatments), as well as time windows and probabilities of carrying out activities. The temporal component also includes weather conditions and vegetation growth models for all vegetation types and crops. Such approach gives a highly realistic, updated on a daily basis, dynamic landscape with vegetation growing in response to the weather, and the pattern of farming activities related to each specific crop, farm, and field. Our methodological framework, supported with semi-automated procedures for spatial data management, makes creating such highly-realistic representations of agricultural landscapes feasible and usable for landscape-scale risk assessment. More importantly, the presented tools allow for testing in silico various scenarios of agricultural practices, including pesticide use, in differently structured landscapes. This seems at the moment the most promising strategy for elaborating sustainable agricultural practices that would allow for high productivity, whilst still protecting the agrobiodiversity. This study was supported by the National Science Centre, Poland (2015/19/B/NZ6/01939).

92 Where are the Springtails? A vertical distribution model for Collembolans V. Roozen, RWTH Aachen University / Institute for Environmental Research / Institute for Environmental Research BioV; L.S. Tschopp, RWTH Aachen University / Institute for Environmental Research BioV; T. Preuss, Bayer Ag / Environmental Safety; A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics; M. Ross-Nickoll, RWTH Aachen
University, Institute for Environmental Research / Institute for Environmental Research

With respect to environmental risk assessment it is crucial to know where and when to protect an organism but still little is known on the dispersal of collembolan communities in agricultural landscapes. Especially for the environmental risk assessment of plant protection products vertical movements can be relevant for exposure assessment of in-soil organisms. Thus, ecological modeling offers a powerful tool to link exposure and effect. We will present the individual-based model of the soil-dwelling collembolan Folsomia candida FOLCAS (Folsomia candida simulation). FOLCAS is a vertical distribution model simulating an agricultural soil column, which can be applied to demonstrate the effect of variations in environmental parameters on the population and its dispersal. In addition, the model features the option to evaluate the effect of a pesticide application. The model consists of two submodels: the lifecycle and the movement submodel. The movement of the individuals in FOLCAS is influenced by temperature, pore space, pH and the organic matter as a proxy for food availability. In order to assess the importance of food availability as a main trigger for movement a vertical distribution experiment was designed. In this experiment we assessed the vertical dispersal of F. candida in OECD soil in relation to food location and time. Transparent PVC columns were filled with 350 g OECD soil up to 20 cm column height and 86 individuals of F. candida of different age classes were added. Each column was divided in 6 compartments from top to bottom: 0-1 cm, 1-2.5 cm, 2.5-5 cm, 5-10 cm, 10-15 cm and 15-20 cm. The location of feeding was varied by different regimes while all other parameters were kept constant (25°C, 60% RH). The results showed that the vertical movement submodel and simulation results of the vertical dispersal of collembolans will be presented. A case study will be used to elucidate the importance of the vertical dispersal of non-target arthropods in effect assessment.

93 A practical application of an individual-based stickleback model in the ERA of PPPs

K. Mintram, University of Exeter / Biosciences; C. Tyler, University of Exeter / Biosciences College of Life and Environmental Sciences; S. Maynard, AstraZeneca / Safety Health and the Environment; A. Brown, Exeter University / Biosciences; C. Liu, Syngenta Weymouth Laboratory; P. Thorbek, Syngenta / Environmental Safety; S. Parker, Cefas Weymouth Laboratory; P. Thobek, Syngenta / Environmental Safety; S. Maynard, AstraZeneca / Safety Health and the Environment. Population models are employed in the environmental risk assessment (ERA) of chemicals, including plant protection products (PPPs), to extrapolate from individual-level effects to predictions of effects on whole populations. Individual-based models (IBMs) allow for the incorporation of individual variability, population-level interaction and specific behaviours. IBMs can therefore be used to extrapolate from a large number of individual-level endpoints and simulate potential effects on populations under realistic environmental conditions. We present an IBM of the three-spined stickleback (Gasterosteus aculeatus) developed for the purpose of predicting population-level effects for exposure to chemicals. The IBM was developed from a series of sub-models parameterised from field data obtained from FOLCAS modelling. Modelled population dynamics (e.g. size/age class distributions and annual fluctuations in population abundance) emerge from the adaptive traits, behaviours and interactions between individuals and their environment (including toxicant exposure). Here, we describe the development and validation of a stickleback IBM and demonstrate its practical application in ERA. Empirical data quantifying the reproductive effects and subsequent recovery, following exposure to a fungicide, were input into the IBM as a sub-model. Various exposure and recovery scenarios were simulated to predict population-level effects over time. The modelled outputs demonstrated that exposure duration and individual recovery post-exposure can influence the overall effects of chemical exposure on population abundance. We suggest that using IBMs to incorporate realistic exposure and recovery scenarios may improve current ERA and result in more realistic protection standards for wild populations.

94 Using the Bayesian network relative risk model to integrate molecular effects, ecological context and ecosystem services to estimate risk over space and time

W. Blanchard, Western Washington University / Institute of Environmental Toxicology; J.D. Stark, Washington State University / Dept of Entomology; K. von Stockelberg, NEK Associates LTD / Department of Environmental Health; C. Mitchell, Washington State University / School of the Environment; V. Chu, Western Washington University / Environmental Science; M. Harris, Whatcom Conservation District / Institute of Environmental Toxicology; L. Wallis, Western Washington University / Institute of Environmental Toxicology. An ongoing dilemma in risk assessment is the perceived difficulty in successfully integrating scales that range from the molecular to ecological, timeframes from days to decades, and endpoints that can be species specific to a host of ecosystem services. Starting in the late 2000s to now there has been an interest in defining ecosystem services and in the calculation of risk to these properties. It has been suggested that ecosystem services are a method to encourage a systems approach to sustainability. Human well-being has become part of the lexicon to included endpoints such as a sense of place, education, employment, public safety and traditional activities. In a recent publication (Harris et al. 2017) it was demonstrated that it is possible to estimate risk in a contaminated site to ecological endpoints, human health and ecosystem services using a clearly defined causal pathways and Bayesian networks. Now we are extending the integration of ecological endpoints, ecosystem services and human well-being from the scale of a contaminated site to that of the Salish Sea. The Salish Sea is a term applied to both the Puget Sound and its watersheds in the United States and the Straits of Georgia in Canada. Vancouver, Seattle, Tacoma, major ports, numerous refineries, paper mills, and high tech industries. The same area is also noted for intense agricultural use, outdoor recreation and the harvest of marine resources. The region is also home to more than 30 recognized Tribes in the U. S. segment and First Nations in Canada. We will use three watersheds in this region, the Skagit, the Nooksack and the Cedar as case studies. Time frames will be from current conditions to 2070 and will include climate change projections into the temperature and precipitation. We will demonstrate the application of the Bayesian-network relative risk model to integrate pesticide effects at the molecular level and the alteration of watersheds to calculate risk to the ecological endpoint Chinook Salmon, the specific economic ecosystem services provide by the endpoint and the watersheds, and finally demonstrate the risks to human well-being as defined from a variety of cultural perspectives.

The environment as a reactor determining fate and toxicity of nanomaterials (II)

95 Mobilisation of silver sulphide nanoparticles in soil column by earthworms’ bioturbation

M. Baccaro, Wageningen University / Toxicology Department; H.H. van den Berg, Wageningen University / Toxicology Department; D. Hermans, L. Sloot, Wageningen University and Research; M. van den Brink, Wageningen University / Dept of Toxicology. Ag2S-NP is the main product of transformation of Ag-NP in the waste water treatment plant (WWTP). In many European countries, sewage sludge containing Ag2S-NPs is applied on the surface of the soils as soil amendments. Earthworms are important ecological engineers in the soil ecosystem, which, on one hand, may be affected by Ag released from the amendments and, on the other hand, may influence the distribution of metals. Therefore, the aim of this study was to determine effects of Ag2S-NP application on an important earthworm driven process, i.e. bioturbation and the effect of the earthworm activity on the vertical distribution of Ag2S-NP in the top soil. Their interplay was assessed in two experiments, in presence or absence of artificial rain fall. Around 2 cm of soil treated with 10 ng Ag kg-1 dry weight soil of Ag2S-NP (28.0±20.4 nm) was applied on top of natural soil columns (10 cm) mimicking an application of 200 Mg sludge ha-1 dry weight. For the first experiment, columns were prepared with and without Lumbricus rubellus and with and without Ag2S-NPs. Every week for 28 days earthworms and four different layers of the soil columns (0-2, 2-4, 6-8, 10-12 cm depth) were sampled. In the same way a second experiment was performed with daily application of 2 nm of artificial rain water, allowing collection of leakage samples from the bottom of the columns. Total Ag content was measured in all samples by ICP-MS following acid digestion and nano-Ag in leakage samples by spICP-MS. Results of the first experiment show that mobility of Ag along the soil column is significantly higher in the columns with earthworms overall. Ag reached the bottom layer of the columns where worms were present while no Ag was found at the bottom layer of the column without worms. This indicates that earthworms do not avoid the contaminated top layer. Ag content in earthworms was relatively constant overtime with an average value of 1.06±0.32 mg Ag kg-1 dry weight. The first study shows that uptake of Ag2S-NPs in earthworms occurred regardless of the partial exposure and points towards the crucial role of earthworm bioturbation in the mobilisation of metal nanoparticles in the top soil. The second experiment of the study is currently being performed, results will be presented at the meeting.

96 Short- and long-term approaches to determine the fate of silver nanoparticles in aqua systems

Engineered silver nanoparticles (Ag ENP) are present in many consumer products. Hence, the ENP enter into sewers and wastewater treatment plants with a high predicted removal into the sludge. If the sludge is applied to agricultural soils, decomposition might result in resuspension of the ENP. The fate and impact of Ag ENP in soils is still unclear. Short- and long-term column remobilization experiments with disturbed soils, short-term column percolation experiments with undisturbed soils, and long-term field lysimeter experiments were conducted. All experiments were performed with sterically stabilized Ag ENP (AgNM-300k), and a slightly loamy Cambisol (RefSoil 01A). Additionally, a medium clayey silt (Luvisol) was used for the column experiments. The column remobilization of the Ag concentration after digestion (Ag\text{digg}) was on a very low level in all percolation steps in both soils. The first percolation step after three days of the Cambisol induced the highest remobilization of Ag which was between 26% and 46% of the Ag\text{digg} concentrations in the soil columns. The correlation between remobilized Ag\text{digg} and Ag\text{digg} concentrations suggests that the remobilized amount of Ag was associated to soil colloids. The breakthrough of Ag ENP in the column percolation experiments was high but incomplete in the Cambisol and the Luvisol. Particularly, columns with preferential flow pathways showed low Ag ENP remobilization. In the unsaturated experiments, a nearly complete retention was found for the Luvisol that showed a clearly smaller pore size structure than the Cambisol. The horizontal displacement of Ag\text{digg} in the lysimeter experiments was low and very likely related to soil tillage as well as bioturbation. A low Ag\text{digg} release to the percolate water (t= 480 d, control= 24 ng l\textsuperscript{-1}, Lysimeter (7 mg kg\textsuperscript{-1}) = 56 ng l\textsuperscript{-1}, DIN 38402-11) was obtained for the lysimeter with the highest Ag ENP application. Ag\text{digg} was determined in a read-out upstream and downstream of the lysimeter in the lower Ag ENP concentration. All roots (wheat, canola, barley) showed a low uptake of Ag\text{digg}. All approaches showed a more or less high retention of Ag ENP in soils why soils are a sink for Ag ENP. However, the remobilization in the lysimeter was incomplete because of root uptake and inhibition of the soil microflora. Thus, the impact of a repeated sludge application to the soil microbiome and the root uptake (e.g. beet) needs further long-term investigations.

97 Determination of attachment efficiency (α) for ENPs in different types of soils by saturated column experiments

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The attachment efficiency (α) has been suggested as the most appropriate fate descriptor for transport of engineered nanoparticles (ENPs) in soils and saturated column experiments as the most accurate method to obtain α. Due to the complexity of the soil composition and texture, a small change in composition or performance of the column protocol may affect the resulting attachment efficiency obtained from the results. The aim of this work is to study the effect of soil composition, flow velocity, initial ENP concentration and the size of ENPs on the calculated attachment efficiency for the specific ENP-soil systems. The α values for nominally 20 and 80 nm citrate coated gold ENPs (Au ENPs), as well as 30 nm sulphonated silver ENPs (Ag\text{S ENPs}) were determined in a read-out upstream and downstream of the column in different soils sampled in the UK. Artificial rainwater was used as the eluent. 10 mM Na\textsubscript{2}CO\textsubscript{3} was used as a conservative tracer to estimate the effective porosity and dispersivity. All columns were packed with an excess of rainwater to limit the amount of air present in the saturated soil. α was either calculated from breakthrough curves of Au/Ag or from the irreversible attachment rate modelled using Hydrus 1D or the relative recovery of the ENPs in the break through curves. Preliminary results show no significant differences in α values for 80 nm and 20 nm Au ENPs. However, the Au ENP breakthrough curves appeared dependent on the flow rate. Even though the shape of the break through curves changes with flow rate, this can be compensated during modeling arriving at consistent α values between the systems with varying flow rates. Furthermore, presence of air in the column affects the distribution of ENPs in the packed columns. Finally, an increase in initial ENP concentration gives higher α values and cannot be accounted for in the equations used for estimating the attachment efficiency. In conclusion, when varying the initial ENP concentration into the columns, the α value is significantly affected. Hence, low NP concentrations need to be used in the column experiments to minimize the reproducibility of calculated α values. Moreover, inclusion of air in the systems appears to induce artefacts that complicate determination of α for specific NP-soil combinations.

98 The transformation of copper and zinc (-nanoparticles) during sewage sludge combustion

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Engineered nanoparticles in wastewater streams are effectively retained by wastewater treatment plants and accumulate in sewage sludge. Digested sludge is subsequently combusted for further volume reduction to allow for phosphorous recovery at a later stage. This study focuses on two metals Cu and Zn, as both are present in digested sludge but are also used as nanomaterials. We investigated (i) the transformation of ZnO and CuO-NP during anaerobic digestion, (ii) the subsequent transformation of Cu and Zn during sewage sludge combustion, and (iii) whether the form of Cu and Zn affects the fate during anaerobic digestion and combustion. We spiked CuO-NP, ZnO-NP, Cu\textsuperscript{2+} and Zn\textsuperscript{2+} to four aliquots of sewage sludge starvation during anaerobic digestion at concentrations for 24h. One aliquot was kept as control. Thereafter, sludge was combusted in a pilot fluidized bed reactor and ashes were collected. Sludge and ashes were prepared for Cu- and Zn K-edge X-ray absorption spectroscopy (XAS) measurements. The speciation of the metals was obtained through linear combination fitting (LCF) of X-ray absorption near edges spectra (XANES) and extended X-ray absorption fine structure (EXAFS) data. LCF fits of experimental XANES and EXAFS data suggest a high degree of sulfidation of both Cu and Zn during anaerobic digestion, irrespective of the spiked form of Cu and Zn. For control sludge and sludge spiked with Zn\textsuperscript{2+}, LCF results from EXAFS data suggest that ~90% of the Zn was present as sulfides, the remaining fraction was best described by a ZnO reference spectrum. For sludge spiked with ZnO-NP, a lower degree of sulfidation (~83% ZnS) with a concomitantly high fraction of ZnO (17%) was calculated. After combustion, EXAFS spectra of Zn were best described by a spectrum of ZnO that was co-precipitated with Ferrihydrite. All LCF fits of Cu in the digested sludge indicated complete sulfidation of Cu. After combustion, LCF fits of the experimental EXAFS spectra revealed the presence of ~30% Chalcopyrite, indicating that Cu was not completely oxidised during the combustion. Comparable findings were obtained for Cu\textsuperscript{2+} and CuO-NP. Further, EXAFS analysis of the sludge and the ashes were very comparable and independent of the added form of Cu. For Zn, however, the addition of ZnO-NP resulted in a slightly lower degree of sulfidation compared to the control sludge and to the sludge that was spiked with dissolved Zn\textsuperscript{2+}. All Zn spectra of the ashes were comparable.

99 Soil ecotoxicity and dissolution of a marketed nanosilver product - a direct comparison with ionic silver

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As part of the REACH Substance Evaluation for silver, new data was generated to further justify read-across from ionic silver to silver nanoforms. Therefore, the soil ecotoxicity and dissolution of ionic silver vs nanosilver were tested. The smallest silver nanoform with the highest specific surface area registered under REACH was used for testing (aqueous suspension containing approximately 37% nanoparticles, degree of sulfidation (~83% ZnS)) with a concomitantly high fraction of ZnO (17%) was calculated. After combustion, EXAFS spectra of Zn were best described by a spectrum of Ag that was co-precipitated with Ferrihydrite. All LCF fits of Cu in the digested sludge indicated complete sulfidation of Cu. After combustion, LCF fits of the experimental EXAFS spectra revealed the presence of ~30% Chalcopyrite, indicating that Cu was not completely oxidised during the combustion. Comparable findings were obtained for Cu\textsuperscript{2+} and CuO-NP. Further, EXAFS analysis of the sludge and the ashes were very comparable and independent of the added form of Cu. For Zn, however, the addition of ZnO-NP resulted in a slightly lower degree of sulfidation compared to the control sludge and to the sludge that was spiked with dissolved Zn\textsuperscript{2+}. All Zn spectra of the ashes were comparable.

100 Tackling nanoparticle fate assessment in surface waters - heteroaggregation as a key process

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The increasing use of engineered nanoparticles (ENPs) inevitably entails emissions to the environment, raising calls for nano-specific environmental risk assessment approaches and regulations. As surface waters are the major receiving compartment, assessing risks requires understanding the aquatic fate of ENPs which, unless soluble, is governed by aggregation, including homo- and heteroaggregation with natural suspended particulate matter (SPM), or stabilisation by natural organic matter (NOM). Due to the omnipresence and larger size of SPM, heteroaggregation is much more likely than homoaggregation. However, integration of this process into fate models and exposure assessment requires parametrisation and is still limited by the lack of simple, yet environmentally relevant experimental protocols. Such could be developed along the lines of the recently adopted OECD testing guideline 318 on ENP dispersion stability, currently accounting only for homoaggregation. The principles of homo- and heteroaggregation are basically the same: the probability of particle attachment is
controlled by the intrinsic particle properties and modified by the hydrochemical conditions (pH, electrolytes, NOM). Distinct from heteroaggregation is the complexity added to the system by SPM in the case of heteroaggregation. In this contribution we therefore propose an approach to develop a heteroaggregation testing protocol based on the OECD TG 318, with a focus on tackling SPM analogues selection. The development of such a protocol requires (1) selecting SPM analogues and heteroaggregation conditions complex enough to represent relevant environmental characteristics, and simple enough for routine testing, (2) an easy-to-handle experimental setup to estimate a heteroaggregation parameter, and (3) an accurate experimental method to validate the latter. Point (1) requires informed simplifications based on a profound understanding of the system. Relevant hydrochemical testing conditions have been established for heteroaggregation in the OECD TG 318 and will also apply for heteroaggregation. However, suitable analogues for natural SPM still need to be selected. We therefore reviewed literature for typical compositions of riverine SPM and carried out screening tests aiming at the creation of complex analogues representing relevant characteristics. Comparisons with simple SPM analogues revealed distinct aggregation behaviour, indicating the importance of complex SPM analogues for heteroaggregation.

**Advances in environmental risk assessment of oil spills and offshore oil & gas operations (II)**

**101** MC-252 biomarkers as indicators of oil exposure and pollutant concentration in sediments of the northern Gulf of Mexico

L.M. Basirico, Louisiana State University; R.J. Portier, Louisiana State University / Environmental Sciences

Different types of crude oil can be identified by the arrangement of constituents, or their chemical fingerprint. In addition, chemical fingerprinting can be used to associate contaminated sediments with specific spill events like the Deepwater Horizon disaster of 2010. Mississippi Canyon-252 (MC-252) source oil, the type of crude oil specific to the Deepwater Horizon event, has been fully characterized by researchers at LSU, including pattern identification of the ion 217 and 218 hance/sterane biomarker families. The presence of biomarkers in the sediments allows for the identification of MC-252 crude oil intrusion into sampled areas, as the arrangement of the ions delineates the source of the quantified PAHs. From 2012 to 2014, five inshore and three offshore transects representing the major estuarine and shauling regions of the Mississippi River delta were sampled to measure sediment concentrations of polycyclic aromatic hydrocarbons (PAHs) and MC-252 biomarkers. Pattern A, pattern B or NO pattern was assigned to each sample based on a visual assessment of the chromatographic profiles. Additionally, a main effects-model was implemented in order to determine the impact of environmental variables, including the presence and pattern of MC-252 biomarkers, on the sediment concentrations of ten PAHs and three toxicity indicators. Ninety-three percent of all sampled sediments (N=1,032) did not contain MC-252 biomarkers. Of the sediment samples containing detectable crude oil biomarkers, 5 percent displayed pattern A and 2 percent, pattern B. Most of the samples containing the families of biomarkers were located at the southern end of the Barataria Bay transect, a region that experienced moderate to heavy oiling during the Deepwater Horizon oil spill event. Additionally, MC-252 biomarkers did not account for any of the variability in the concentrations of the measured pollutants across the study. A main effects model was used for the analysis. The lack of MC-252 biomarkers in the vast majority of the sampled sediments, indicated that the PAH contamination in the current study was not from the Deepwater Horizon oil spill event. There is need for better markers of the origination of PAHs in freshwater and marine sediments. Furthermore, crude oil is not the only or even the best indicator of potential toxicity of these sediments.

**102** Downregulation of hsp90 and increased intermoult duration in the blue crab, Callinectes sapidus, in response to oil exposure

S. Chiasson, Loyola University / EEB; S.M. Giltz, C.M. Taylor, Tulane University / Ecology & Evolutionary Biology.

The 2010 Deepwater Horizon (DWH) oil spill in the northern Gulf of Mexico (NGOM) resulted in over 780 million liters of crude oil spilling into Gulf waters. In an effort to disperse the oil, nearly 7.6 million liters of dispersant was applied. In response to oil exposure Callinectes sapidus, in response to oil exposure

**103** Physiological and molecular impacts of crude oil and/or dispersant-contaminated seawater and sediments on the sponge Halichondria panicea (phylum Porifera).

J. Vagh, Heriot-Watt University / School of Energy, Geosciences, Infrastructure and Society; J.M. Roberts, The University of Edinburgh / Grant Institute; T.B. Henry, Heriot-Watt University / The School of Energy, Geoscience, Infrastructure and Society.

Sponges (phylum Porifera) are a diverse group of filter-feeder organisms present in most aquatic environments. In the marine environment, sponges perform a wide range of ecological functions including cycling of nutrients such as carbon, nitrogen and silica; and, in areas where they are present at high densities (spoon grounds), they provide a habitat for a diverse range of benthic organisms. Because of their importance within marine ecosystems, the impacts of anthropogenic activities such as hydrocarbon exploration and production on marine sponges must be assessed. The objectives of this study were to: (1) determine the physiological impact of crude oil and/or dispersant contaminated seawater and sediments in model sponge Halichondria panicea; and (2) characterise the effects of crude oil and/or dispersant contaminated seawater exposure on the transcriptome of H. panicea. A series of 48-hycyclics aromatic hydrocarbon (252)-containing the families of biomarkers were located at the southern end of the Barataria Bay transect, a region that experienced moderate to heavy oiling during the Deepwater Horizon event, has been fully characterized by researchers at LSU, including pattern identification of the ion 217 and 218 hance/sterane biomarker families. The presence of biomarkers in the sediments allows for the identification of MC-252 crude oil intrusion into sampled areas, as the arrangement of the ions delineates the source of the quantified PAHs. From 2012 to 2014, five inshore and three offshore transects representing the major estuarine and shauling regions of the Mississippi River delta were sampled to measure sediment concentrations of polycyclic aromatic hydrocarbons (PAHs) and MC-252 biomarkers. Pattern A, pattern B or NO pattern was assigned to each sample based on a visual assessment of the chromatographic profiles. Additionally, a main effects-model was implemented in order to determine the impact of environmental variables, including the presence and pattern of MC-252 biomarkers, on the sediment concentrations of ten PAHs and three toxicity indicators. Ninety-three percent of all sampled sediments (N=1,032) did not contain MC-252 biomarkers. Of the sediment samples containing detectable crude oil biomarkers, 5 percent displayed pattern A and 2 percent, pattern B. Most of the samples containing the families of biomarkers were located at the southern end of the Barataria Bay transect, a region that experienced moderate to heavy oiling during the Deepwater Horizon oil spill event. Additionally, MC-252 biomarkers did not account for any of the variability in the concentrations of the measured pollutants across the study. A main effects model was used for the analysis. The lack of MC-252 biomarkers in the vast majority of the sampled sediments, indicated that the PAH contamination in the current study was not from the Deepwater Horizon oil spill event. There is need for better markers of the origination of PAHs in freshwater and marine sediments. Furthermore, crude oil is not the only or even the best indicator of potential toxicity of these sediments.

**104** Advances in the effects of UV on oil toxicity in aquatic organisms

A.P. Roberts, K. Bridges, University of North Texas / Advanced Environmental Research Institute; J. Morris, Abt Associates; B.J. Venables, University of North Texas / Advanced Environmental Research Institute; M.O. Krasne, Abt Associates; M.L. Gielazyn, NOAA / co USEPA Region IV

The 2010 Deepwater Horizon (DWH) oil spill in the northern Gulf of Mexico (NGOM) resulted in over 780 million liters of crude oil spilling into Gulf waters. In an effort to disperse the oil, nearly 7.6 million liters of dispersant was applied. In response to oil exposure

**105** Photoenhanced Toxicity of Petroleum to Aquatic Invertebrates and Fish: Review of the Science

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Photoenhanced toxicity is a distinct mechanism of petroleum toxicity that is mediated by the interaction of solar radiation with specific polycyclic aromatic compounds (PACs) in oil. Photoxicity is observed as a 2 to greater than 1000-fold increase in chemical toxicity to aquatic organisms that have also been exposed to light sources containing sufficient quantity and quality of ultraviolet radiation (UV) [1]. While there is a long history of petrogenic pollution, photoenhanced toxicity has only been reported for sun-weathered middle distillates, crude and heavy oils can exhibit photoenhanced toxicity. These same products do not exhibit photoxicity in standard test protocols because of low UV irradiance in laboratory lighting. Fresh water, estuarine and marine waters have been shown to have sufficient solar radiation exposure to elicit photoenhanced toxicity, and a diversity of aquatic invertebrate and fish species can exhibit photoenhanced toxicity when exposed to combinations of oil and UV. Risks of photoenhanced toxicity will be greatest to early life stages of aquatic organisms that are translucent to UV and that inhabit the photic zone of the water column and intertidal areas exposed to oil.

Fish model species in human and environmental toxicology (II)

Life-stage, and species-specific effects of dietary methylmercury exposure
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Mercury is a globally distributed contaminant, found even in remote aquatic ecosystems. Once deposition occurs, it can be microbially transformed into organic forms, such as methylmercury (MeHg) [1]. MeHg is highly bioavailable, and it bioaccumulates and biomagnifies in biota leading to potentially toxic body burdens in long-lived organisms at high trophic levels. MeHg can be actively transferred from mother to offspring, through large amino acid transporters with the potential to cause severe, irreversible effects on developing organisms. Here, we describe the development loss in offspring of seawater-adapted and oil-adapted MeHg-exposed fish in future detergent production in teleosts has been established. The bidirectional communication between the gut and the central nervous system (CNS) is referred to as the gut-brain axis, which plays an important role in behavior, brain function, neurodevelopment, and the progression of neurodegenerative disorders. Therefore, we characterized MeHg-mediated changes to the gut microbiome composition in P. promelas adults. Because the dopaminergic system is highly conserved among taxa, we sought to confirm the altered dopamine concentrations in P. promelas brains in a higher vertebrate species. Metabolomics was performed on the mid-brains of male mice (Mus musculus CD-1) exposed to similar concentrations of dietary MeHg for 30-days. Changes in dopamine concentrations of the telost brain were mirrored in the mid-brains of male mice, and several other significant changes to the mouse mid-brain metabolome were detected. Collectively, these results suggest current environmental exposure scenarios to MeHg are sufficient to induce a number of molecular-level changes that are associated with costs to whole organism fitness, with consequences for multiple life stages, and species. Due to the similar changes detected in mice, there is increasing evidence to suggest teleosts as a surrogate model species for studies assessing effects of MeHg on highly conserved systems in higher vertebrates.

Characterization of molecular toxicity pathways of Fluoxetine in rainbow trout and white sturgeon using RNA-Seq whole transcriptome analyses
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The increasing number of emerging chemical contaminants (ECCs) and their unknown effects to aquatic ecosystems serves as an impetus to develop advanced environmental risk assessment (ERA) approaches to improve regulatory decision-making. This is because current ERA rely on live animal testing that are expensive, time consuming and of ethical concern. Furthermore, the use of model organisms does not assure protection towards native species because of the limited understanding of their mode of action, and of their variable sensitivity to pollutants. Hence, there is need to establish an unbiased approach to characterize toxicity pathways that allow probing of an entire biological system without a priori knowledge of the mechanisms of toxicity. Advances in ’omics technologies can improve current testing strategies as they offer high-throughput and cost-effective approaches to examine patterns of mechanistic toxicity which could guide endpoint selection and mechanistic pathway model development. As a result, objective characterization of ecotoxicity pathway models to predict outcomes of regulatory relevance for the selective serotonin reuptake inhibitor, fluoxetine (FLX), in 2 fish species of concern in Canada. Juvenile rainbow trout (RBT) and white sturgeon (WS) were exposed to 125 μg/L FLX in 9th static-renewal system, and sequence-by-synthesis whole transcriptome analysis was used to determined unique and differential expression in fish livers and brains. A 0.05 cut-off false discovery rate identified differentially expressed contigs between FLX and control groups. A total of 406 and 429 contigs were significantly altered in RBT livers and brains, respectively. Of these, 238(59%) and 236(55%) matched unique gene names. In WS, 252 and 192 contigs were significantly altered in livers and brains, respectively, with 145(58%) matched unique gene names. Pathway analysis using ontologies based on zebrafish in KEGG and GO Consortium showed a total of 101 affected pathways. Over half(58%) of the affected pathways were involved in biological processes while others were involved in cellular components (13%), molecular function (18%), and genetic information processing (11%). The results of this study will be compared to apical outcomes assessed in a parallel chronic study, and which will allow the assembly of toxicity pathways across multiple levels of organization with the end goal of identifying molecular markers that are indicative of adverse effects.

Transgenerational effects of early life stage exposure to endocrine disruptors across biological scales in a euryhaline model fish
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Emerging research demonstrates that EDCs, which agonize, antagonize, and/or disrupt endocrine activity in model species, such as Menidia beryllina, a euryhaline fish with short generation time that is found throughout North America and is demonstrated to be sensitive to contaminants. As such, we exposed Menidia beryllina embryos (8 hpf) until 21 dph to environmentally relevant of an androgenic or estrogenic EDC of emerging concern: levonorgestrel (Levo) (10 ng/L), bishydroxynitrin (Bi) (5 ng/L), respectively, and coupled this exposure with testing of an established androgenic or estrogenic EDC: trenbolone (TB) (10 ng/L), and ethinylestradiol (EE2) (5 ng/L). We are now evaluating the potential for transgenerational EDC effects across three generations, with EDC exposure isolated to the parental generation (to 21 dph) only, across biological scales. This study is examining changes in gene expression, DNA methylation, histological analysis of reproductive organs, as well as altered fecundity, sex ratio, morphology, and immune response in the F0, F1, and F2. We are also sequencing the M. beryllina genome. F0 results show that early-life exposure to EE2 significantly skewed adult sex ratios (feminized) relative to controls. Findings from the F0 and
Sustainable Development Goals: the global context defining the agenda for government, business and academia

Can the Agenda 2030 and the Sustainable Development Goals be the drivers to change the world? E. Giovanni, A.SviS

How the SDGs are being addressed in Horizon 2020 M. Tamborra, European Commission - DG Research and Innovation

Examples of EU projects related to SDGs M. Recchiioni, European Commission - EASME

Why SDGs are relevant for a large enterprise A. Valcalda, ENEL

Conclusions E. Tonda, UN Environment / Division of Technology, Industry and Economics (DTIE)

Questions and answers

Mercury Biogeosciences - Fate, Effects and Policy

Rethinking Atmospheric Mercury Chemistry M. Gustin, University of Nevada, Reno / Natural Resources and Environmental Science

Mercury (Hg) is considered a global pollutant. This is because it has a long atmospheric residence time. Because of the continued and increasing emissions of this pollutant to the atmosphere associated with anthropogenic activities, and the fact that once released from a geologic repository an atom of Hg may be potentially bioavailable for thousands of years, the Minamata Convention was developed and has come into force. This global treaty focuses on protecting human health and the environment from the adverse effects of mercury. There are 3 general forms in the atmosphere- gaseous elemental Hg, gaseous oxidized Hg (Hg(II) or Hg(III) compounds), and that bound to particles. Gaseous elemental Hg can be transformed to gaseous oxidized Hg (GOM) by a variety of atmospheric oxidizers. Once generated, GOM is readily deposited to ecosystems. Understanding the chemistry of GOM is important for predicting deposition velocities, availability in ecosystems, and potential for conversion to methylmercury. Methylmercury is a subtle neurotoxin and is bio-accumulated in ecosystems. Recent work using cation exchange membranes in the University of Nevada Reno –Reactive Mercury Active System (UNR-RMAS), and an air Hg calibrator system (Utah State University) have demonstrated that the standard measurement method for GOM –collection on a KCl denuder- results in underestimation of GOM concentrations by 2- to 13 times. In addition, thermal desorption profiles of GOM compounds collected using nylon membranes indicate that different chemical forms exist in the atmosphere. Data collected in urban areas, in the marine boundary layer, and at high elevation indicate that GOM compounds present are influenced by oxidants present in the air.

100 Integrated OMICS and imaging for a better understanding of ecotoxicological mechanisms - PAH developmental toxicity as an example E. Velmurugan, A. Bhadra, Department of Physical Sciences, Earth and Environment; R.O. Barra, Universidad de Concepcion / Aquatic systems; J. Gavilán, Universidad de Concepcion / Biomarcadores; S. Casini, University of Siena / Scienze Fisiche della Terra e dell’Ambiente; M. Fossi, University of Siena / Department of Physical Sciences, Earth and Environment; R.O. Barra, Universidad de Concepcion / Cellular Biology, Faculty of Biological Science

The BioBio River (Central Chile) is the third most important basin in Chile and is considered a Biodiversity Hot Spot with 17 native species. It presents a high degree of intervention by the convergence of point and diffuse contamination of complex chemical mixture and establishes the possible relationships between the physiological/reproductive alterations observed and the high degree of intervention of this river.

112 Poster spotlight: MO248, MO249, MO256

111 Physiological / Reproductive Status of Native Fish Exposed to a Complex Chemical Mixture in the BioBio River, Central Chile M. Quirolo-Zara, Universidad de concepcion / Biomarcadores; S. Casini, University of Siena / Scienze Fisiche della Terra e dell’Ambiente; M. Fossi, University of Siena / Department of Physical Sciences, Earth and Environment; R.O. Barra, Universidad de concepcion / Aquatic systems; J. Gavilán, Universidad de concepcion / Cellular Biology, Faculty of Biological Science

The BioBio River (Central Chile) is the third most important basin in Chile and is considered a Biodiversity Hot Spot with 17 native species. It presents a high degree of intervention by the convergence of point and diffuse contamination of complex chemical mixture and establishes the possible relationships between the physiological/reproductive alterations observed and the high degree of intervention of this river.

114 How different stages of development of the oocytes determined for this species (n = 2332, p < 0.05). On the other hand, the collected specimens show a difference in weight and length, presenting specimens of less frequency of length in the lower third with respect to those present in areas with less intervention. These responses are associated with the increase of the values of environmental parameters towards this zone. The results of this study indicate a gradient of adverse biological effects by the convergence of point and diffuse contamination of complex chemical mixture and establishes the possible relationships between the physiological/reproductive alterations observed and the high degree of intervention of this river.
Different oxidized forms are produced in the free troposphere, marine boundary layer, and due to local oxidants in urban areas. Understanding atmospheric chemistry of GOM is important for developing instruments that will accurately measure GOM, and helping guide policymakers in developing solutions for reducing Hg emissions and contamination of ecosystems.

120 Evaluating spatial dynamics and species variation on mercury and selenium molar ratios in Northeast Atlantic marine fish communities

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Seafood is the main dietary source of methylmercury (MeHg) exposure for humans and MeHg is a primary contaminant of concern for seafood consumption advisories. Co-occurrence of the Selenium (Se) and mercury (Hg) in seafood directly affect their bioavailability and toxicity. The protective and antagonistic effects of Se against Hg have been assumed to be due to Se inhibition of MMHg formation. This study was designed to assess the antagonistic effect of Se against Hg in marine fish samples collected from the Barents Sea, Norwegian Sea, North Sea, Skagerrak, North Atlantic and Norwegian fjords and coastal areas between 2006-2015. The mean Se levels ranged from 0.27 to 0.78 mg kg⁻¹ wet weight (ww) in Atlantic cod (Gadus morhua) and wolffish (Anarhichas spp.), respectively. The mean Hg levels ranged from 0.04 to 0.77 mg kg⁻¹ ww with the lowest level in Atlantic mackerel (Scomber scombrus) and the highest in blue ling (Molva diphrygea), leading to variation in the mean Se:Hg molar ratios from 1.9 in blue ling to 43.2 in Atlantic mackerel with Hg levels contributing most to the variation. In general, pelagic species had the lowest Hg levels and the highest Se: Hg molar ratios whereas deepwater demersal species had the highest Hg levels and the lowest Se: Hg ratios. Most species had a large portion (more than 50%) of specimens with a Se: Hg ratio exceeding 5 except for tusk (Brosme brosme) (4% less than 1, 53% between 1 and 5) and blue ling (19% less than 1, 80% between 1 and 5). Se and Hg levels showed weak positive correlation (R from 0.24 to 0.70) in 13 of 17 species. The Se: Hg ratio was negatively correlated to fish length and Hg levels. Mean Se: Hg molar ratios varied across offshore areas for all species and a gradual decreasing trend was found for all species from north to south due to increasing Hg concentrations. The EU maximum level of Hg and the Se: Hg molar ratio above one, we emphasize that fish from NEAO are generally safe regarding Hg contamination. Less than 1% of the total analyzed specimens had Se: Hg molar ratio less than 1 and the surplus Se may ameliorate the toxic effect of Hg to some extent.

121 The interaction of mercury and selenium across environmental media

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Both mercury (Hg) and selenium (Se) can bioaccumulate within aquatic ecosystems and are toxic to organisms when found at high concentrations. Individually, high concentrations of Hg can affect water and biota, but the presence of both trace metals has been suggested to have antagonistic effects. Due to this relationship, many studies propose that increased environmental concentrations and consumption of Se is a pathway to reduce Hg toxicity in organisms. Yet, despite this important link, little is understood about the biogeochemical processes that promote this antagonistic relationship. In fact, a large amount of research has been conducted to understand these processes, but the understanding of how these processes occur in marine systems is limited. In this study, we seek to better understand the uptake of Hg and Se into biota, as well as the biogeochemical conditions that promote this pathway and evidence for antagonistic effects. We use samples collected from the mountaintop mined region of West Virginia, USA, where high concentrations of contaminants have previously been found in these watersheds. To answer this research question, we analyze total Hg (THg), MeHg, total Se (TSe), and Se precipitation in water, sediment, biofilm, stream macroinvertebrate, and spider samples. Our results show that Hg, MeHg, and Se are bioaccumulating in the food chain, with the highest concentrations found in macroinvertebrates. We also find that the concentration of Se is an important factor that limits MeHg bioaccumulation and has the potential to reduce the bioavailability of Hg in biota, thereby reducing its bioaccumulation. This is especially important in oligotrophic and nutrient-limited systems, where Hg concentrations are lower, and the concentration of Se is higher.

Constraining Uncertainties in the Global Mass Balance of Mercury Using Observations and a Bayesian approach

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Uncertainties in global mercury mass balance are constrained in this work using all currently available observations of mercury species in the global environment, and a previously published multimedia model for mercury, using WinMOM3. Reducing uncertainties helps in estimating mercury concentrations and mass balances with greater confidence. Ten key input parameters that were identified to be significantly contributing to the output uncertainties in previous studies. These included: emissions of mercury to the atmosphere, reduction and oxidation of mercury in surface and sub-surface oceans, and partition coefficients of mercury species groups (Hg(0), Hg(II), Hg-p) in surface and sub-surface oceans. Then, a survey of literature on observations of mercury in the global environment is made. As these observations (for example, concentration of total mercury in air) are also key model outputs, we can update model inputs by comparing model simulated outputs to the actual observations. For this updating, a Markov chain Monte Carlo (MCMC) technique called Metropolis Hastings which is based on the Bayes rule is adopted. The observed concentrations of Hg(0) in atmosphere, dissolved gaseous mercury, and total mercury in surface ocean are collected from published literature and used to obtain a likelihood function. Input parameters and their confidence range are revised. A revised mass balance is obtained through a forward Monte Carlo analysis using updated inputs. It is found that the uncertainties in key input parameters (such as partitioning of reducible divalent mercury between suspended solids and water in surface oceans) have been constrained to a considerable extent, explaining the likelihoods’ sensitivity to about 50% for these parameters. In all, the uncertainties in key output results, such as evasion of mercury from ocean to atmosphere, are also reduced. From factor 650 to factor 50, and in net reduction of Hg(II) to Hg(0) in atmosphere from factor 36 to 10. Analysis of contribution to variances of inputs to output variances suggests that, still, parameters describing oceanic processes such as partitioning between suspended solids and water, and mercury redox biogeochemical reactions contribute more to variances in key model outputs, compared to parameters such as global mercury emissions. Therefore, more significant effort must be made in understanding mercury process in oceans rather than in conducting emission inventory exercises.

123 Effects of probable nutrient limitation on the relationship between mercury and marine microorganisms in seawater

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Mercury redox reactions in oceans, contribute strongly to mercury outputs, compared to parameters such as global mercury emissions. Therefore, more significant effort must be made in understanding mercury process in oceans rather than in conducting emission inventory exercises.

Bioavailability and realistic risk assessment of organic contaminants
125 Anisotropic exchange kinetics of organic contaminants with passive samplers in stagnant sediment: is multiple-thickness passive sampling the better alternative?

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Passive sampling with thin polymer sheets is increasingly recognized as a superior release method for both hydrophobic and nonpolar organic chemicals in sediment porewater. For deducing true freely dissolved concentrations in the aqueous phase from measured polymer concentrations, the compounds are required to reach thermodynamic equilibrium between the polymer and the water phase. However, for in-situ deployment in stagnant sediment equilibration times are beyond practical time scales. The spiking of passive samplers with performance reference compounds (PRCs) has therefore been introduced as a way to deduce equilibrium concentrations from the release of PRCs over the deployment period. This approach relies on the assumption of isotropic exchange kinetics between the uptake of the native compounds into the polymer and the release of spiked PRCs from the polymer. Our aim was to test whether this assumption is valid in stagnant sediments in in-situ and ex-situ conditions, considering different types of sediment and various spiked concentrations. For the field study, we immersed low-density polyethylene (PE) and silicone thin sheet passive samplers of multiple thicknesses and spiked with PRCs for 5 months into contaminated sediment in the Oslo harbour. For the ex-situ study, Oslo harbour sediment and Hortex harbour sediment was incubated with PRC-spiked PE under stagnant conditions at room temperature in the laboratory and samples were taken at various time points. From the PRC depletion field data, sediment porewater concentrations were modelled using the Fernandez-one-dimensional-diffusion model. In the additional field, equilibrium concentrations were modelled using a multiple thickness one-dimensional diffusion model. The ex-situ uptake and release data were modelled with a one-dimensional diffusion model (uptake) and a simple exponential one-compartment model (release). This was done to test whether both in-situ data and release data for uptake and release kinetics were not identical. In addition, the ex-situ experiment revealed that PRC release kinetics is also dependent on the initial PRC spiking concentration and in conclusion, the data question the usefulness of PRCs for passive sampling in sediment, as the use of polymers of multiple thicknesses can produce results that are free from biases caused by anisotropic exchange kinetics.

126 Sediment toxicity of chlorpyrifos: whole sediment bioassay vs. silicon disc passive dosing

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Realistic risk assessment of sediments polluted with organic contaminants is much more complex than tests with water samples. Physical and biological processes as well as contaminant bioavailability, may strongly influence the adverse effects on test organisms in whole sediment testing. The current work is the first series of studies to our group on whole sediment-equilibrated silicon rubber (ESR) could allow for a major simplification of the assessment of the overall impact of organic contaminants in sediment. The aim of the current study was to demonstrate that ESR can transfer the chemical activity of the insecticide chlorpyrifos from spiked sediment to aquatic bioassay with ESR as a passive dosing material. The effect level of chlorpyrifos in a 28d whole sediment bioassay was compared to effect levels observed in a 4d ESR passive dosing test using first instar larvae of the midge *Chironomus riparius*. Additional sampling with polycarbonate solid phase microextraction (SPME) fibers in both sediment and ESR dosed water was used to align the bioavailable concentrations in both tests designs. The ESR samplers accumulated chlorpyrifos up to 12% of the total spiked chlorpyrifos amount within 1 month. SPME samplers in sediment had 1-3x lower concentrations than SPME equilibrated with ESR. Thus, the chemical activity in sediment as well as that released from the ESR in water were comparable within a factor of 3. The insecticide chlorpyrifos showed only slightly more toxic effect levels in a 28d whole sediment test than in the 4d ESR dosing assay. Bioavailable concentrations in SPME samples in both assays indicated leathally toxic freely dissolved concentrations in the range of 0.2 - 0.1 pg/L. This study suggests that the 4d ESR dosing assay with sensitive first instar midge larvae provides valuable and realistic insight in the toxic potency of insecticide contaminated sediment comparable to much more elaborate 28d whole sediment tests. Also, at lowest tested toxic insecticide levels, concentrations in SMPE extracts were close to detection limits, so accurate measurements of safe bioavailable chlorpyrifos concentrations via SPME becomes problematic. This suggests that ESR dosing assays and chronic whole sediment studies could be used more effectively to demonstrate specific pollutant toxicity than chemical analysis of realistic sediment exposure levels.

127 Implementing desorption extraction methods into bioavailability-oriented bioremediation

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Regulators are starting to consider bioavailability within retrospective risk assessment frameworks for organic chemicals, however, implementation is not straightforward because the development of bioavailability science have not always been translated into ready-to-use approaches for regulators. Possible pathways for translating bioavailability science into regulation of organic chemicals have recently been identified (Environ. Sci. Technol. 49:10255–10264, 2015). A simplified approach was proposed in which the assessments of soil/sediment and their contaminants should be based on two measurable values: the total extractable concentration and the bioavailable concentration as measured with robust and reproducible chemical and/or biological methods. One of the chemical methods which has been proposed to measure bioavailability of hydrophobic chemicals (HOCs) such as PAHs is the desorption extraction with Tenax during 20 h (ISO 17402) (Environ. Toxicol. Chem. 20:706–711, 2001; Integr Environ Assess Manag, 11:208–220, 2015). Understanding the role of bioavailability in the biodegradation of chemicals is relevant not only for retrospective contaminated land management but also prospective risk assessment applied in the approval and regulation of organic chemicals. With the aim of providing pathways for implementation into regulatory contexts, we carried out desorption extraction measurements with Tenax in a greenhouse experiment in which different strategies (use of soil/sediment plots PAHs from 6 years at least 25 years) were used, soil, oil, water and soil/sediment concentrations was modelled using the Fernandez-one-dimensional-diffusion model. In addition, the field equilibrium concentrations were modelled using a multiple thickness one-dimensional diffusion model. The ex-situ uptake and release data were modelled with a one-dimensional diffusion model (uptake) and a simple exponential one-compartment model (release). This was done to test whether both in-situ data and release data for uptake and release kinetics were identical. In conclusion, the data question the usefulness of PRCs for passive sampling in sediment, as the use of polymers of multiple thicknesses can produce results that are free from biases caused by anisotropic exchange kinetics.
contaminated soils collected from two gasworks sites in the UK. Site 1 had a total polycyclic aromatic hydrocarbons (PAHs) concentration of 3500 mg/kg, and total aliphatic concentration of 1500 mg/kg. Site 2 had a lower concentration of both aromatic and aliphatic hydrocarbons 250 mg/kg and 350 mg/kg respectively. Heavy metals (HMs) concentrations in both soils ranged between 600 and 1200 mg/kg and the main elements were Zn, Pb and Cu. Both soils were amended with either 5% biochar or 15% compost in order to achieve metal stabilization and enhance soil degradation. The total uptake of available contaminant concentrations of the selected fractions as well as HMs were determined after 0, 30, 90 and 180 days. Further to this microbial biomass, soil respiration, phospholipid analysis, Microtox toxicity, earthworm’s lethality and seeds germination were carried out to assess how ecological health changed as the soils underwent remediation treatments. The study showed that, for both soils, microcosms amended with compost showed the most significant reductions in toxicity. The microbial number and respiration activity increased by two orders of magnitude after compost addition. Conversely, there was no significant difference between non-treated and biochar amended soils. At the onset of the experiment, no seed germination was observed in Soil 1 (non-treated) whereas an increase in seed germination was observed after 90 days for mustard and 30 days after 30 days for peas. The compost treatments had the highest percentage germination for both soils across all seed types. Similarly, the earthworms assay showed there was significantly greater survival rate at the end of the experiment compared to the onset. The biochar treatment resulted in a lower survival counts compared with compost treatment, with the non-treated samples having the lowest results. Preliminary results suggest that addition of compost and biochar accelerated the degradation rate of hydrocarbons component and contributed in reducing toxicity of the soils. The degradation of PAHs and re-distribution of HMs will be evaluated with a multivariate analysis; data will be explored to highlight associations between contaminant’s concentration (reduction) and its influence on biological properties and toxicological responses in mixed contaminated soils.

Impact of Biochar Additions to Soil on Contaminant Sorption and Plant Bioavailability

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Reclaimed water is increasingly used in arid and semi-arid regions for irrigation. Contaminants in the reclaimed water, (e.g. pharmaceuticals and personal care products (PPCPs) and metals) could accumulate in exposed crops. Biochar is a potentially cost-effective soil amendment and contaminant sorbent that could reduce the plant bioavailability of reclaimed water associated contaminants. But is biochar truly effective when plants come into contact with contaminated soils? Do plants undergo pyrolysis (decomposition at high temperatures with no oxygen)? The main objective of this study is to investigate the impact of wood biochar on the crop bioavailability of selected PPCPs often found in reclaimed water. A secondary objective is to quantify the contaminant sorption-desorption characteristics in the amended soils and to determine if there is a relationship with plant bioavailability. PPCPs were selected as target contaminants because of their widespread occurrence in reclaimed water and their potential impact on animals feeding on the irrigated crops. Target PPCPs were selected based on chemical properties, widespread use, frequent detection in WWTP effluent, and potential risk to the environment. The target PPCPs represent a range of therapeutic uses including antibiotics linked to antibiotic resistance in bacteria (sulfamethoxazole (SMZ) and trimethoprim (TR)), an anticonvulsant that prevents seizures and relieves nerve pain (carbamazepine), and an antidepressant (fluoxetine (FLX)), and an antihyperlipidemic (gemfibrozil (GBZ)). Atrazine (ATZ) was also selected because it is an herbicide commonly used on corn and has been used in a number of sorption studies with biochar. Pinyon Juniper, Russian Olive, and Lodgepole Pine derived biochars were chosen because they have the lowest results. Preliminary results suggest that addition of compost and biochar amendments to the soils significantly increased seed germination of mustard and rye grass, and after 30 days for peas. The compost treatments had the highest percentage germination for both soils across all seed types. Similarly, the earthworms assay showed there was significantly greater survival rate at the end of the experiment compared to the onset. The biochar treatment resulted in a lower survival counts compared with compost treatment, with the non-treated samples having the lowest results. Preliminary results suggest that addition of compost and biochar accelerated the degradation rate of hydrocarbons component and contributed in reducing toxicity of the soils. The degradation of PAHs and re-distribution of HMs will be evaluated with a multivariate analysis; data will be explored to highlight associations between contaminant’s concentration (reduction) and its influence on biological properties and toxicological responses in mixed contaminated soils.

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A novel framework for a new generation of water consumption indicators in LCA and footprint studies

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Towards global regionalized characterization factors for water consumption impacts on instream freshwater ecosystems

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Available freshwater consumption indicators for use in LCA and water footprint studies implemented in LCA software are based on either a volumetric approach of the water consumed (what we called first generation indicators) or on stress indicators what we called indication of indicator. The first generation only performs an inventory and thus a partial footprint assessment according to ISO 14046, while the second generation shows limitations in describing the consequences of a lack of water at the endpoint. To date, no LCA method comprehensively distinguishes water masses (e.g. surface water and ground water) and water transport flows (e.g. inter-basin transport) within the boundaries of a watershed (e.g. evaporation) and beyond (e.g. groundwater and air advection), thus overlooking details in hydrological processes that affect environmental relevance of the assessment. In addition, a structured LCA framework is currently lacking, as can be observed by the scattered and often incompatible developments of water fate and impact assessment models published in recent years. These models are all valuable contributions in themselves, but impossible to combine to an integrative global characterization model that makes such developments operational in LCA. The challenge of improving environmental relevance of current water consumption indicators has been tackled by the Water Use in LCA (WULCA) working group within the UN Environment Life Cycle Initiative by developing consensus-based guidelines for the third generation of water consumption indicators, with a focus on consistency of water used, contamination and environmental relevance. The unified framework is also available, approaches, the new generation of indicators will include, for the first time, and fate and transport of water flows in a global regionalized multimedia model. In the framework, we provided spatial and temporal specifications for the LCA and specified data requirements for the LCI. To demonstrate how to apply principles and recommendations of the guidelines, we developed an illustrative example. The operationalisation of the guidelines has the potential to harmonize current and future methods under a unique framework and to enhance the environmental relevance of the water use impact category in LCA.
quantify potential water consumption impacts on freshwater ecosystems. In existing mechanistic models, mostly based on species-discharge and species-area relationships, ecological requirements are not taken into account. This implies considering equal response to stress for different taxa, overlooking the relationships between species and their habitat, along with other aspects of biodiversity (e.g. abundance). In this study we want to show the importance of habitat modelling to describe the impact path of water consumption on ecosystem quality. We propose a new approach to incorporate habitat effects and discuss the applicability at the regional and global scale. Water consumption may alter stream discharge and other related physical variables. Habitat suitability equations can be used to quantify physical habitat availability for freshwater species in Weighted Usable Area (WUA). Starting from WUA equations, a Habitat Characteristics Potential model is proposed for river fish species. The model of invertebrates is proposed and applied in France at Q50 (wet season) and Q90 (dry season) flows. HCP represents the change of available habitat area deriving from river discharge alteration. At the river reach scale, HCPs from different taxa have been aggregated under different perspectives in order to test the results’ sensitivity to negative and positive effects of hydrological alteration. A spatial aggregation has been still tested in the work of Alkire and sub-watershed. Subsequently, the global HCP model’s applicability has been discussed. HCP is highly correlated with river size. The aggregation at reach scale is driven by specific taxa and by positive HCP scores (habitat loss). The result of the aggregation at watershed is consistent with existing evaluations of hydromorphological pressures in Europe. The main challenge in applying HCP globally is due to hydrological and hydraulic data availability. It is more likely to find comparable European and extra-European species habitat preferences. The proposed model is a promising effect factor for mechanistic impact characterisation which should be integrated with fate factor models describing hydrological alteration at a comparable spatial resolution. Since habitat models are based on species abundance, HCP represents the first step towards developing biodiversity damage indicators complementary to species loss.

134 The use of dynamic stock model to the definition of characterisation factors for biotic resources depletion
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Biotic natural resources have received little attention by the LCA community and this tempers the use of LCA for fish based food and feed products. Current LCA methods do not assess the impact of biotic stocks depletion. The global biotic stock (i.e. population) is a well-studied topic in theoretical ecology and it is commonly used for fish stocks management. The main of the present work is the use of this knowledge to assess biotic resources depletion in LCA. This is illustrated by the definition of characterisation factors (CF) for fish stocks depletion. The relationship between inventory and impact is often quantified as marginal change in impact according to a marginal change in inventory. Here, the Depleted Stock Fraction (DSF) is considered to be the assessed impact while the elementary flow of the inventory is a mass of fish removed from the biomass stock. The marginal approach is applied to the Schaefer model, a commonly used model in fisheries representing the dynamics of the biotic stocks. It combines catches, current biomass and maximum intrinsic growth rates of the stock (population). To determine these parameters for most of the world fisheries, catch time series were used for species of FAO Global Fishery and Aquaculture Statistics. This allows assessing all fisheries of the world in the LCA framework with a midpoint biotic resource depletion impact. To our knowledge, this work is one of the first assessments of depletion in LCA, based on a model of population dynamics. We are confident this will bring improvements to the LCA of fish-based products, allowing for the comparison of different fishery alternatives with respect to the use of marine biotic resources. One of the extensions of this present work could be the definition of CFs for terrestrial biotic resources where similar dynamic stock models are used. This advocates the use of a similar approach based on population model dynamics for both terrestrial and marine biotic resources.

135 Accounting for soil quality effects of agricultural land management in LCA
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The growing demand for food and feed has put a pressure on the agricultural sector, which resulted in agricultural intensification. This entails many adverse environmental impacts and takes a huge toll on land resources; which can result in land degradation. Especially farm management can affect soil quality (e.g. soil organic carbon, SOC) by for instance the choice of crop rotations. The agricultural sector is therefore forced into a more sustainable approach. To assess the environmental sustainability, life cycle analysis (LCA) is a powerful tool. As soil quality is an inherent aspect of agricultural sustainability, life cycle impact assessment (LCIA) methods should account for the effect of agricultural land management on soil quality. Despite a lot of efforts that have already been made, there are still several research needs on the integration of soil quality effects in LCIA methods. Ideally, inventory data are translated into the effect on the environment by indicators moving along the cause-effect chain from mid- to endpoint towards an area of protection (AoP). Although soil quality changes are often related to the AoP ecosystem health, we focus on the AoP natural resources as we refer to the impact of land management on soil by the long-term ability to produce biomass. To improve the usefulness in the agricultural sector, different land use intensities should be distinguished. We therefore introduce three interdependent LCIA indicators. At early midpoint, SOC changes (SOC1; indicator 1) are used to indicate the long-term effect of agricultural land use on this. This can result in biomass productivity losses (BPL, indicator 2). At endpoint, we propose additional land requirements (ALR, indicator 3) as indicator, which corresponds to the area needed to produce the yield that has been lost. To calculate characterization factors (CFs), we chose as reference situation the highest achievable SOC stock and yield, which are calculated for each initial soil quality stock. The models RothC and EU-Rotate are used to quantify SOC stocks and yields, respectively. CFs are developed for several rotation systems in Flanders. Though, the elaborated framework is generally applicable and allows the calculation of CFs for other regions. Thanks to the use of an achievable reference situation and a distinction between land management strategies (good, bad and standard practices), the indicators can be a useful tool to strive for a more sustainable agriculture.

136 Poster spotlight: MO093, MO094, MO106

Environmental fate of emerging contaminants in the water cycle: analytical challenges and engineered solutions (II)

137 Full scale WWTP balancing with passive samplers offers new insights in xenobiotic elimination processes
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Calculating elimination rates for full scale wastewater treatment plants is very demanding because it requires the knowledge of mixing regimen to match inflow to out flow volumes which translates very often in sampling campaigns that span over several days. While a certain progress in understanding elimination processes of xenobiotics has been gained from laboratory reactor tests, reliable and large datasets from real-life full scale plants are still missing due to the cumbersome sampling effort. Here we implemented the use of passive samplers to establish a clear parameter table to find correlating marginal and extra-European species habitat preferences. The proposed model is a promising effect factor for mechanistic impact characterisation which should be integrated with fate factor models describing hydrological alteration at a comparable spatial resolution. Since habitat models are based on species abundance, HCP represents the first step towards developing biodiversity damage indicators complementary to species loss.

138 Screening of wastewater-borne pharmaceuticals and their phototransformation products in rivers
S. Lopez, N. Montemurro, IDAEA-CSIC / Environmental Chemistry; D. Barcelo, IQAB-CSIC / Department of Environmental Chemistry
Pharmaceuticals are continuously discharged into the rivers from wastewater treatment plants and hence be able to directly calculate elimination rates. The method was validated in a pre-study with parallel autosampling and then applied to 18 WWTPs representing a large range of design properties such as hydraulic and sludge retention times. Normalisation with carbamazepine and lidocaine proved to be robust since both inlet-outlet ratios were well correlated and elimination rates of the investigated compounds fell into the ranges documented in literature. Furthermore it was possible to identify patterns of elimination by applying a cluster analysis and several compound elimination rates were found to negatively correlate with sludge retention time and hence to be more related to active biomass of the sludge. Inlet loads of the compound could be calculated by calibrating the more invariant outlet concentrations to passive sampler masses and then back calculating to inlet loads via the elimination rates. Population equivalent loads proved to be within expected ranges from the literature and non-domestic sources could be identified. Passive sampling might hence close the gap of investigation in xenobiotic behaviour on full-scale treatment plants and serve as well as a routine performance surveillance tool.

138 Screening of wastewater-borne pharmaceuticals and their phototransformation products in rivers
S. Lopez, N. Montemurro, IDAEA-CSIC / Environmental Chemistry; D. Barcelo, IQAB-CSIC / Department of Environmental Chemistry
Pharmaceuticals are continuously discharged into the rivers from wastewater treatment plants. Hundreds of wastewater-borne pharmaceuticals have been detected in river samples but their concentrations along the river change constantly due to additional inputs and natural attenuation processes. Apart from biodegradation, drugs can undergo phototransformation reactions by either direct or indirect photolysis including reaction with oxygen (O2), hydroxyl radical (·OH), peroxyl radicals (‘ROO'), photo-excited organic matter, and other reactive species. To evaluate these processes in a river, usually laboratory studies are
performed in a first stage and then in the next step studies are conducted directly in the natural environment. In our group, we proposed a workflow using the combination of HRMS and processing software for evaluating the phototransformation of pharmaceuticals on a single compound basis under simulated and real environmental conditions. In contrast to this compound-by-compound approach, in the new approach presented here, degradation was not assessed for a single compound but instead a cocktail of human drugs was subject to the transformation processes. Following the identification of photo-TPs, a list of suspect TPs was created and used to screen them in SPE-concentrated river water samples. For the generation of photo-TPs, reconstructed surface water was spiked with a cocktail of 34 pharmaceuticals at concentrations of 10 µg/L and exposed to artificial light in a sunlight simulator. Using UPLC-MS/MS (L-generation determined spectrophotometrically and disinfection effect of iodine measured by fcal coliform tests. 0.35 maximum mM iodine concentration could be synthesized during experiments. During iodine production, while phenolic compounds' concentrations were decreased (50% acetaminophen removal in real effluent wastewater treatment plant), removal of non-phenolic compounds such as naphroxen were also observed (50%). The results have shown that the biocatalytic generation of I2 was possible using laccase-mediator system. Iodine production was affected by the initial laccase activity and mediator concentration. Laccase catalyzed bactericial activity in municipal wastewater was also assayed without the addition of any mediator assuming that wastewater already contains mediators such as acetaminophen. Using this system, non-ficollic compounds present in the tested wastewater were removed.

### 139 Degradation of a polymer probe exposed to different wastewater environments: Linking chemical transformations and potential microbial consumers

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The objective of the present contribution was to link the chemical transformations observed in a polymer probe exposed to the different aquatic environments found in a WWTP with the microbiological communities present in situ. Phylogenetic composition of free living bacteria was compared to polymer-attached microbial communities. Specifically, chemical degradation and microbial community characterization was carried out in the influent flow (IN), in the secondary aeration tank (ATER), in the effluent (ATER) and some of the photo-TPs was still underway. Several photo-TPs of our database were detected in the extracts of the surface water samples. Thus, this approach highlights that UPLC–HRMS is a powerful tool for qualitative analysis, allowing the search for photo-TPs. With the detection of some photo-TPs we have provided evidence for photolysis and thus underpinning the importance of natural attenuation processes in rivers.

### 140 Optimization of Laccase Catalyzed Iodine Synthesis as Enzyme Based Disinfectant

g. yuksel, Universite de Sherbrooke / Civil Engineering.

In addition to traditional and new generation water pollutants, microbial contamination is still one of the major problems which has affected the potential recycle and reuse of wastewater for domestic or industrial usage. Selection appropriate treatment approach has played an important role not just to reach discharge limits, it is also important to decrease human footprint in the environment. Some hydrolytic and oxidative enzymes (i.e., Laccase) is capable of oxidizing unreactive iodide to reactive iodine, when they can play roles to degrade recalcitrant pollutants in wastewater. The resulting iodine represents a powerful antimicrobial compound. The aim of this study is investigating the potential of acetophenone and phenolic organic contaminant acetylaminophen as mediator in a laccase mediator system to generate disinfectant iodine. The stability of reaction could be synthesized during experiments. During iodine production, while phenolic compounds' concentrations were decreased (50% acetaminophen removal in real effluent wastewater treatment plant), removal of non-phenolic compounds such as naphroxen were also observed (50%). The results have shown that the biocatalytic generation of I2 was possible using laccase-mediator system. Iodine production was affected by the initial laccase activity and mediator concentration. Laccase catalyzed bactericial activity in municipal wastewater was also assayed without the addition of any mediator assuming that wastewater already contains mediators such as acetaminophen. Using this system, non-ficollic compounds present in the tested wastewater were removed.

### 141 Halogenated methanesulfonic acids in drinking water - Identification, standard synthesis, and analysis

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Persistent, bioaccumulative, and toxic (PBT) substances have been monitored since the 1960s. PBT substances might be, among others, pharmaceuticals, personal care products, or industrial chemicals, however, a significant fraction may also be dead-end transformation products (TPs), and thus a substantial share of them might still be unknown. In a non-target screening approach dedicated to the identification of mobile, potentially drinking water relevant organic contaminants, we identified chlorinated and brominated methanesulfonic acids (MSAs) as novel water contaminants and estimated the concentrations of the most prevalent congeners to be in the 100 ng/L range for some drinking water samples. Accurate quantification, however, was hindered by the lack of commercially available reference materials. Thus, we synthesised chloromethanesulfonic acid, dichloromethanesulfonic acid, bromomethanesulfonic acid and bromochloromethanesulfonic acid as well as O3-trifluoromethanesulfonic acid (as internal standard) and included these analytes in a sample pre-treatment and hydrophilic interaction liquid chromatography – tandem mass spectrometry (HILIC-MS/MS) method dedicated to the analysis of very polar water samples taken from high population areas in different countries.

### 142 Poster spotlight: MO272, MO273, MO274

Building of large-scale inventories of emissions and resources and applications for environmental footprints of territories, nations and sectors

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The European Inventory of Existing Commercial chemical Substances (EINECS) lists over 100,000 chemical substances used on the market. Over 16,000 chemical substances have been registered in REACH since 2008. In comparison, only ca. 3,000 energy carrier substances are known from life cycle impact assessment (LCA) to express their potential toxic impact on human health (cancer and non-cancer effects) and freshwater ecosystems. Because of human activities, those pollutants may enter the environment in several different ways: they are emitted to air from the combustion of materials, released through wastewater from industries and households, applied to soils together with manure and pesticides, etc. Combined with the limited availability of release data, the sheer number of substances and the large variety of emission sources are challenges that one needs to overcome to quantify the overall toxic impacts of a country. Here, we therefore propose an updated methodology to build national inventories of toxic emissions in EU Member States in 2000-2014. The framework builds on earlier works and differentiates environmental compartments (air, water and soil) as well as anthropogenic sources (industries, households, manure and pesticides) application on agricultural soils. It relies on existing and publicly available data, and extrapolation techniques are developed and used to fill in the gaps across countries in the entire period 2000-2014. The resulting harmonized inventories cover more than 500 substances, including both organics and inorganics such as persistent organic pollutants and heavy metals. Although still limited in substance coverage, it is thus possible to analyze the contribution of each substance and anthropogenic source to the toxic impacts on human health (human toxicity) and freshwater ecosystems (freshwater ecotoxicity), using LCA methods such as the consensus model USEtox.

144 Combining economic modelling and LCA to assess regional policies: key learning points from a case study on the French forestry sector
T.B. Beaussier, INRA; E. Loiseau, Ifremer; S. Caurla, INRA

Economic modelling is increasingly used in Life Cycle Assessment (LCA) to perform consequential LCA for the environmental assessment of product and services. Economic models can also provide significant enhancements for assessing the effects of regional policies, such as in territorial LCA approaches. Among them, equilibrium models appear as a good compromise to assess both socio-economic and environmental impacts of regional policies in an exhaustive and representative way. However, there are still some bottlenecks when trying to combine both approaches in practice. For instance, the levels of system aggregation and distinction between industry, households, manure and pesticides production is required to ensure a consistent combination while limiting the time spent to collect data. This talk aims at providing insights on the combination of a partial equilibrium model, the French Forest Sector Model (FFSM), and LCA to assess the eco-efficiency of two regional policies supporting local wood industries in the French East Region. Two approaches will be used to combine economic models with LCA - a Life Cycle Inventories (LCI), i.e. i) Extended Environmental Input Output modelling and ii) a method based of MFA (Material Flow Analysis) and process-based LCA. Eco-efficiency ratios based on economic and environmental impacts allow identifying scenarios with best environmental performances. In addition, this combination allows considering supply and demand dynamics, and thus the socio-economic effects of a decision. Using two different approaches, we are able to compare strengths and weaknesses of both types of combinations and discuss them considering policy assessment results, system representation and system boundaries. Thus, our work provides both insights on down-to-ground policy analysis and methodological developments on combining economic modelling with LCA. Here, economic modelling outputs are used as LCA inputs but more integrated modelling could be performed for completeness and optimization purposes. Perspectives on a stronger coupling will also be discussed.

145 A regional life cycle approach for assessing the climate change mitigation potential of bioenergy production
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Keywords: Regional, spatial, biobased economy, GHG While traditional life cycle assessment is a powerful tool, for spatial applications, it is limited. With the ever increasing drive towards regional bio-based circular economies, as a means of ensuring future climate change mitigation, there is a need to produce more regional and spatially representative life cycle assessments of biobased systems and bioeconomy regions. “RELCA”, a Regional Life cycle inventory approach, was developed to assess the regional and spatial variation in the environmental performance of bioenergy production within a focus region. Through the use of catchment delineation, conventional geographical modelling is combined with life cycle software to assess the potential environmental burdens of regional bioenergy configurations (i.e. bioenergy plants and their biomass catchments). RELCA was used to assess the climate mitigation potential of biodiesel, for the region of Central Germany. With this approach we showed, for the focus region, that the mitigation potential changed between the different biodiesel configurations, due to their location within the region. When compared to a fossil diesel comparator (83.8 CO2eq/ MJ), the climate change mitigation potential of the regional biodiesel ranged between 53%-62%. When the results were compared to the typical RED (Renewable Energy Directive) values, a 13-31% greater mitigation potential than the RED was observed. The latter, illustrating that regional variability cannot be captured with a simple regional average value or default value. Additionally, scenarios were used to test the mitigation potential of reduced nitrogen fertiliser application during the production phase. The results indicated that while reduced nitrogen fertiliser led to emission reduction per hectare, the greater land area input required to meet the demand of the biodiesel plants, led to an increase in emissions ranging from 1.14-5.71 g CO2eq/MJ biodiesel. Thus, highlighting that assessments of biobased systems should consider configurations of biomass and conversion plants in order to determine appropriate mitigation strategies. Therefore, the ability to account for the territorial and geographical characteristics found within a region, using life cycle approaches, is important to support more sustainable regional resource management.

146 LCA_WIND_DK: temporally, geographically and technologically-sensitive life cycle inventories for the Danish wind turbine fleet

The environmental performance of a wind turbine is usually calculated as the ratio of the life cycle impacts occurring during the turbine production phase to the electricity it produces during the use phase. The modelling of the life cycle inventory in each phase should ideally cover the temporal, geographical and technological dimensions of the product system under study. Assumptions are commonly used to simplify and handle variable aspects of the inventory. While this approach provides generic, one-size-fit-all inventories, it may disregard important characteristics of the wind turbine leading to biased end-results. As these assumptions are prone to differ from one study to another, the results become hardly comparable. With more than 1,500 wind turbine models on the market and a high variability of sites and manufacture periods of the different installations, it makes the environmental assessment of wind turbine production a daunting task. LCA_WIND_DK: a.dk wind energy life cycle assessment (LCIA) to express their potential toxic impact on human health (cancer and non-cancer effects) and freshwater ecosystems (freshwater ecotoxicity), using LCIA methods such as the consensus model USEtox.

147 Assessing environmental impacts of individual households: A large-scale bottom-up model for Switzerland
A. Froemelt, ETH Zurich; R. Buffat, ETH Zurich / Institute of Cartography and Geoinformation; N. Heeren, S. Hellweg, ETH Zurich / Institute of Environmental Engineering

Besides governmental consumption, household consumption is the main driver of economy, and is thus ultimately responsible for the environmental impacts that occur throughout the whole life cycle of goods and services, including their manufacture, installation, and decommissioning. Therefore, assessing environmental footprints of households is an important basis to identify environmental policies. This study aimed to develop a comprehensive regionalized bottom-up model for Switzerland that is able to assess the environmental impacts induced by individual households. The purpose of this model is to provide a virtual platform for detailed scenario analysis which shall support effective political decision making on different scales. Three existing bottom-up models were merged: a building stock energy model, an agent-based transport simulation and a household consumption model. All of them were tested and evaluated beforehand. The physically-based building energy model establishes
simplified energy balances for each residential building based on spatially and temporally resolved climate data, building characteristics and 3D geometries. It provides estimates of space heating, hot water and electricity demand for each household. The mobility sub-model builds upon the results of an agent-based traffic simulation framework which was applied to Switzerland and reproduces mobility patterns of Swiss inhabitants in space and time. The third sub-model produces a data-driven approach and enables the quantification of consumption of food, consumables, and other goods and services for each Swiss household by means of data mining techniques. Linking these sub-models with environmental background data allowed for computing an environmental profile for each household in Switzerland. The application of this model to the current situation of Switzerland reveals interesting differences between individual households, district heating regions and different scenario assumptions. By covering the variability of household behavior and quantifying the demands and environmental footprints of households within a certain area, the model delivers important insights for local policymakers to derive targeted environmental strategies tailored to the specific problems and household types in a region. Furthermore, the high resolution of all three sub-models permits testing of policies and in-depth analyses of scenarios, ranging from local renewal and refurbishment programs to future mobility solutions such as autonomous vehicle systems.

148 Poster spotlight: MO109, MO110, MO113

Mechanistic effect modelling for risk assessment: applications, use in a regulatory context and future directions

149 Modelling ecological scenarios for the assessment of chemical effects on stream communities

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The ecological risk assessment of chemicals (ERA) aims at quantifying the likelihood of adverse ecological effects posed on populations and the communities they comprise. Effects caused by the exposure of organisms to chemicals can however to a great extent depend on environmental scenarios as well on the states, behaviours and interactions of organisms with consequences for individual life history, population responses and community dynamics. In this regard, our major objective is to suggest how to model stream ecological scenarios for ERA. We suggest to employ ecological classifications as defined within the Water Framework Directive. Here, the ecological scenario is a virtual representation of an ecosystem, which involves both abiotic components (habitat scenario) and biotic components (the functional and life history scenario). Technically, we integrate spatial explicit habitat information in form of raster maps, temporal information on abiotic factors like temperature and chemical exposure, functional trait data bases, dynamic energy budget models and process based effect models to simulate macroinvertebrate and fish assemblage dynamics. In model applications, we explore to what extent the ecological scenario will affect the adverse outcome of chemical exposure.

150 Robust implementation of TKTD models with Bayesian inference

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The application of toxicokinetic-toxicodynamic (TKTD) modeling proved to be of particular interest in strengthening the Environmental Risk Assessment (ERA) of chemicals compounds (e.g., REACH dossier accounting for toxicity of industrial discharge, evaluation of impacts of Plant Protection Products (PPPs), ...). TKTD models describe the time-course of processes leading to toxicity at the level of organisms. These models include all mechanisms from the toxicokinetics part (describing the compound fate from external concentration to internal kinetics (e.g., exposure, uptake, elimination, biotransformation, internal distribution), and translate the internal concentration into toxicodynamics covering alteration of cells and organs functioning that can eventually lead to a toxic effect at the organism level (e.g., mortality, reduced reproduction, abnormal behavior) then affecting the population dynamic. In for survival analysis of organisms in response to a chemical stressor, the Generalized Unified Threshold Model of Survival (GUTS) is today recognized as a suitable and powerful TKTD framework incorporating two complimentary death mechanisms: Stochastic Death (GUTS-SD) and Individual Tolerance (GUTS-IT), from which a large range of existing models can be derived. Under governmental institutions as the OECD have acknowledge the necessity of TKTD models for ERA improvement, but while an integrative mathematical framework as GUTS offers an efficient theoretical approach, its practical use is challenging (from model implementation to parameter estimation), especially with time-variable exposure. The Bayesian approach has multiple advantages as (i) using all data provided by the experiments, (ii) taking into account the knowledge from experts and/or previous studies, (iii) being relevant for complex model with small dataset since there is no degree of freedom, and (iv) a clear handling of uncertainties by providing distributions of parameter posteriors. InTo ease the access of Bayesian fitting of GUTS models based on ordinary differential equations, we compared several implementation of GUTS models. To do so, two software packages were used. The first one, BayesGUTS, incorporated the widespread statistical language R (JAGS and Stan). Then, we embedded those algorithms within two R packages with the core idea to be user friendly (e.g., using experiment design to define priors). A side result is to propose a step-by-step approach to perform Bayesian statistics in ecotoxicology.

151 Can TKTD-models describe and predict synergistic interactions in Chironomus riparius?

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The azole fungicides propiconazole and prochloraz are known to enhance the toxicity of pyrethroid insecticides like α-cypermethrin during co-exposure. The development of these synergistic actions in the waterfly Daphnia magna have recently been modelled using toxicokinetic (TK) and toxicodynamic (TD) models in order to describe the underlying mechanisms for the enhanced toxicity. The purpose of the current study was to predict the same C. riparius TKTD-framework for synergistic interactions in D. magna can be applied to the midge larvae Chironomus riparius to describe development in survival rates and the underlying mechanisms over time when co-exposed to azole fungicides and pyrethroid insecticides. Toxicity of the individual compounds was tested using a pulsed concentration response design with an initial 24 hour exposure period followed by six days of recovery in clean water. To assess the potential of these azoles and α-cypermethrin were a range of tests conducted with co-exposure to 1, 3, 10, 30, or 100 µg L⁻¹ of propiconazole or prochloraz and 2.5, 5.0, or 10.0 µg L⁻¹ of α-cypermethrin. For the TK-modelling will uptake and elimination rates of the individual compounds in C. riparius be measured to parameterize the TK-model before using the internal dosimetry to the observed toxic effects. We hypothesise that the synergistic interactions can be described and modelled by adding a synergy parameter “s” to the biotransformation rate constant for α-cypermethrin and that the value of this “s” parameter will depend on the azole exposure concentration. The preliminary results indicated time-dependent synergistic interactions in C. riparius as previously observed in D. magna, but also a higher sensitivity of C. riparius towards the fungicides with 168 h EC₅₀-values for the 24 h pulse exposure of 1.06 ± 0.27 and 0.28 ± 0.10 µmol L⁻¹ for propiconazole and prochloraz, respectively. This is surprising as previous non-published data indicated that C. riparius has an approximately 10 fold faster initial elimination rate of the azoles compared to D. magna. We expect that our TKTD models will be able to explain these kinetic differences and how they relate to the observed toxicity. We further hope to that model could be used to describe the population dynamics of C. riparius exposed to the pesticide chlorpyrifos and the azoles α-cypermethrin, propiconazole and prochloraz with varying time intervals between the pulses.

152 Integration of temperature-dependent TKTD kinetics in individual-based population modelling - A case study with Chaoborus crystallinus

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The toxicokinetic-toxicodynamic (TKTD) model framework GUTS is increasingly used and becoming the standard effect model in regulatory risk assessment. However, this model is mostly used without the temperature dependency of TKTD kinetics, and effect measurements are usually performed in the laboratory at 20°C. This approach is rather unrealistic, especially in outdoor scenarios with significantly different water temperatures over the year: On the one hand, in cases with low water temperatures during autumn and winter, the toxic effects can be reduced or delayed, while on the other hand, the degradation of the substance is often slowed down, which increases the exposure time. But also at higher temperatures than 20°C, increased toxic effects on organisms are to be expected. In this presentation, the influence of seasonal temperatures on toxicity at the population level is exemplarily examined. For this purpose, an individual-based population model for the phantom midge Chaoborus crystallinus is used. This has been intensively tested using outdoor aquatic mesocosm studies, and has been coupled to the TKTD framework GUTS, which has been extended by a temperature dependency based on laboratory data. Temperature dependencies of lethal effects on C. crystallinus larvae exposed to the pesticide chlorpyrifos and the azoles D. magna were measured in the laboratory in the range of 4-20 °C. These data are used to parameterize the GUTS, and subsequently to simulate the population dynamics of Chaoborus under variable outdoor temperature conditions. The Chaoborus model was used with and without the implemented temperature dependency of the effect model to assess whether toxic effects on a Chaoborus population are increased or decreased depending on the temperature conditions. Therefore representative summer and winter exposure scenarios were selected.
Using temperature dependencies for the relevant biological and toxicological processes, this modelling approach allows a more realistic risk assessment of pesticides for populations in the field.

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**Assessing lethal and sublethal effects from time variable exposure for different life-stages with the DEB model: an example for a Pyriehroid in rainbow trout**

E. Zimmer; IBACON GmbH; T. Preuss, Bayer Ag / Environmental Safety; S. Norman, RidgewayEco; B. Minten, ADAMA Deutschland GmbH; V. Ducrot, Bayer Ag / Environmental Safety

Ecotoxicology

The study investigates effects of beta-cyfluthrin on juvenile rainbow trout (Oncorhynhus mykiss) using TK-TD modelling. As part of the risk assessment modelling is used as a supporting tool to back up the experimental results and as an investigation tool to better understand the mechanisms of effects of beta-cyfluthrin. Beta-cyfluthrin is acting as neurotoxicant in fish for which the severity of effect depends on the magnitude and duration of the exposure peak. To address these characteristics, the effects of beta-cyfluthrin on rainbow trout were evaluated with two independent early life stage tests (ELS): a standard Tier 1 study with constant exposure and a Tier 2c study under time variable exposure. Observed effects differed in these two studies. Under constant exposure, severe mortality and significant growth effects were observed while under peak exposure, no effects on survival were observed, and only negligible effects on growth were found. The model was successfully calibrated using the constant exposure experiment, and then accurately predicted the effects observed in the peak-exposure assay. The model was capable of simulating from the start to the end of the experiment the observed differences in the effects of beta-cyfluthrin a) on early life stages of the fish.

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**Atmospheric Microplastic’s: A novel method for the identification of microplastic’s in the inhalable size range.**

Using temperature dependencies for the relevant biological and toxicological processes, this modelling approach allows a more realistic risk assessment of pesticides for populations in the field.

New Horizons in Particulate Polymer Analysis: Micro- and Nanoplastics and Tire Rubber Detection, Characterisation and Impacts in the Environment

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**Prediction of effects on chemicals on three-spined stickleback populations in mesocosms**

V. David, INERIS; B. Goussen, University of York / Environment; J. Porcher, INERIS / INERIS UMRI SEBIO ECOT; R. Beaudouin, INERIS / Models for Ecotoxicology and the internal METO

To improve environmental risk assessment, mechanistic models predicting the impacts of toxpoints on populations such as individual-based models (IBM) was suggested as relevant tools. Furthermore, IBMs can be coupled with DEB (Dynamic Energy Budget) models which describe physiological processes of an organism. However, the development of DEB-IBMs requires a large number of data on the organism’s physiology, which make them difficult to build. To this aim, data from mesocosm experiments can be of great interest for developing and calibrating DEB-IBMs. One of the species that can be used in mesocosm experiments is the three-spined stickleback (Gasterosteus aculeatus). Furthermore, the ecology and biology of this telost fish is relatively well-known and a DEB model for this organism has already been developed. In this study, we used data from several mesocosm experiments to describe stickleback populations under control conditions, and exposed to three concentrations of an endocrine disruptor, the Bisphenol A (BPA, 1, 10 and 100 μg/L). First, using two sets of experiments in control conditions, different ways of integrated temperature and food data was tested in order to assess the relevance of the DEB model calibrated with laboratory data for stickleback populations (population size, male, female and juvenile frequencies, lengths and coefficient of variations) were well described and were used to compare with the endpoints of mesocosms exposed to BPA. In conclusion, simulated endpoints of stickleback populations can thus be used as a baseline to compare exposed populations to BPA in order to improve environmental risk assessment. In a second step, the DEB-IBM could be adapted in order to introduce the effects of toxpoints such as BPA on the individuals and thus extrapolate the effects at the population level.

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**Analysis of polystyrene based microplastics in the environment**

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Marine anthropogenic litter is a severe environmental problem. Wastes discarded or deposited in aquatic environments (including rivers, estuaries and coasts) are usually mixed with other pollutants being the 80% of these plastics. One of the main issues is the extreme stability of plastic wastes. Under environmental conditions, the erosion of these materials generates smaller fragments some of them in the nano- and microscopic scale, which are known as nanoplastics (NPLs) and microplastics (MPLs), respectively. The quantitative analysis of these plastic debris is particularly difficult because of their physicochemical properties (low solubility, a wide range of molecular weights, etc.) and potential contaminations sources in the laboratory. For these reasons, different approaches should be considered to find a standardised protocol for the determination of MPLs and NPLs in the environment. In this context, this study was focused on the investigation and practical comparison and combination of different analytical tools for the quantitative and qualitative analysis of MPLs/NPLs using: (1) techniques to assess the physicochemical properties such as Thermogravimetric Analysis or TGA, Differential Scanning Calorimetry or DSC, and Fourier-Transformed Infrared Spectroscopy or FT-IR; (2) quantitative and qualitative information by techniques based on direct mass spectrometric as high resolution mass spectrometry with Electrospray ionization (ESI), Atmospheric Pressure Chemical Ionization (APCI), Matrix-assisted Laser Desorption Ionization (MALDI), Desorption Electrospray Ionization (DESI) and Direct Analysis Real-Time (DART). These studies have been carried out using as a representative polymer the polystyrene (PS), which is one of the most frequently used for plastics production. Finally, LC-APPI-HRMS complemented by other techniques such as TGA, DSC and FT-IR allowed obtaining qualitative and quantitative information about of the whole spectrum of polymers, which may be present in the environment.

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**Uptake, egestion and accumulation of microplastic in mussel after an experimental exposure**

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Filter feeding invertebrates such as mussels are especially susceptible target species to microplastic (MP) ingestion. Field and laboratory studies have reported that MP are ingested by mussel. Once ingested, MP may be egested through defecation,

SETAC Europe 28th Annual Meeting Abstract Book
Tire wear particles (TRWP) as environmental contaminants have received interest since the 1960s [1]. TRWP have adverse effects on human health [2]. TRWP can be regarded as microplastics, because the rubber component of TRWP is mainly (natural and synthetic). With regional differences, the contribution of TRWP to the microplastic emissions to the environment can reach up to 60% [3]. Analysis of TRWP is challenging because of the high variance in composition. Results showed that after 120 hours of the exposure the 6% and 2% of the TRWP ingested was accumulated in the digestive gland of mussels exposed to the Low and High MP dose, respectively. The diameter of this MP (around 3 μm) was significantly lower than that of the MP offered (8 μm) and the MP ejected (6-9 μm). This suggested a specific removal through faeces of larger MP particles and the retention of smaller ones in the digestive system.

158 Analysis of tire wear particles in environmental samples using TED-GC-MS

160 Are we speaking the same language? Towards a definition and categorization framework for environmental plastic debris
M. Wagner, Norwegian University of Science and Technology / Department of Biology; N.B. Hartmann, Technical University of Denmark / DTU Environment; A. Verschoor, RIVM / Centre for Safety of Substances and Products; T. Hüffer, University of Vienna / Department of Environmental Geosciences; M. Hassellöv, University of Gothenburg / Department of Marine Sciences; R.C. Thompson, Plymouth University / School of Marine Science and Engineering

Research and Testing / 5.3 Mechanics of Polymers; M. Albrect, TU Chemnitz; T. Reemtsma, Helmholtz Centre for Environmental Research / Department Analytical Chemistry.

In this contribution we present the analytical method development based on Zn and S content and apply the analytical method to determine tire wear particle concentrations along the treatment path of road runoff. Tire wear particles have been recognized as an important environmental pollutant. Analytical methods for the quantification of tire wear particles in environmental samples are still under development and struggle with multiple sources or insufficient stability of markers. We developed an analytical method which allows quantifying tire wear particles in road runoff, sediments and surface waters. Tire wear particle quantification is based on elemental composition and distinct elemental ratios. The analytical method aims at i) tire wear particle enrichment using density separation followed by ii) a stepwise assisted acid digestion and elemental detection of Zn and S using ICP-MS and ICP-OES. Elemental content of the particulate fraction in the water samples was analysed after filtration only, since the amount of solids was too low for the density separation procedure. Acknowledgement - The authors thank the BMBF for funding the MiWa project (reference number 02WRS1378H) and BWB for provision of samples.
pelagic and reef fishes, including mahi-mahi (*Coryphaena hippurus*) and bicolour damselfish (*Stegastes partitus*). To date, within the marine teleost group, nothing is known about how crude oil exposure affects the detection of olfactory cues or if crude oil can be detected and avoided. To address these questions, the time that control and oil-exposed bicolour damselfish spent in a chemical alarm cue and the time that control and oil-exposed mahi-mahi spent in diluted crude oil was examined using a two-channel flame response system. Exposure to undiluted crude oil availed a consistent chemical alarm cue, whereas exposed conspecifics did not avoid the cue (p < 0.001). Control mahi-mahi did not distinguish between seawater and crude oil, however, oil exposed mahi-mahi spent a greater proportion of time in crude oil than the control fish (p < 0.01). Moving forward, an electro-olfactogram technique will be used to measure the generator potential from the olfactory epithelium of bicolour damselfish and mahi-mahi to detect the response to olfactory cues following oil exposure. This research was made possible by a grant from The Gulf of Mexico Research Initiative. Grant No: SA-1520; Name: Relationship of Effects of Cardiac Outcomes in fish for Validation of Ecological Risk (RECOVER).

162 A wide range of endpoints are impacted by oil exposure in early and later life stages of marine fish M. Grosell, J.D. Stiegelitz, RSMAS University of Miami / Marine Biology and Ecology; R.M. Heuer, University of Miami / Marine Biology and Ecology; C. Pasparakis, Rosenstiel School of Marine Sciences / Marine Biology and Fisheries; L. Schneider, RST, CAS Oiled Fish. Such reductions in cardiomyocyte contractility are likely related to impaired cellular calcium cycling also suggested by RNAseq data. This research was made possible by a grant from The Gulf of Mexico Research Initiative. Grant No: SA-1520; Name: Relationship of Effects of Cardiac Outcomes in fish for Validation of Ecological Risk (RECOVER).

163 Investigating the endocrine disruptive and genotoxic potential of crude oil samples using adapted in vitro toxicity tests S. Johanna, Institute for Environmental Research RWTH Aachen University; L. Nuesser, RWTH Aachen University, Institute for Environmental Research / Department of Ecosystem Analysis; L. Spencer, Institute for Environmental Research RWTH Aachen University / Department of Ecosystem Analysis; D. Altin, BioTrax; H. Hollett, RWTH Aachen University / Institute for Environmental Research; T. Seiler, RWTH Aachen University / Ecosystem Analysis

In case of an acute oil spill the decision on optimal response techniques to achieve the best environmental benefit is based on important knowledge about regional ecosystem function. Even adult marine fish are sensitive to brief, low-level oil exposures, showing reduced aerobic capacity and reduced performance, reduced maximal oxygen uptake, and reduced visual acuity in later stage juveniles. Juvenile fish exposed to oil show altered olfactory responses, reduced prey capture ability, and higher susceptibility to predation, likely due to altered central nervous system function. Even adult marine fish are sensitive to brief, low-level oil exposures, showing reduced aerobicscope and swimming performance. The lower swim performance in adult oil-exposed fish with a normally developed heart, is due to reduced cardiac output driven by reduced stroke volume. Work on isolated cardiomyocytes show that sarcomere shortening upon electrical stimulation is reduced by acute oil exposure which likely explains the reduced stroke volume observed in vivo. Such reductions in cardiomyocyte contractility are likely related to impaired cellular calcium cycling also suggested by RNAseq data. This research was made possible by a grant from The Gulf of Mexico Research Initiative. Grant No: SA-1520; Name: Relationship of Effects of Cardiac Outcomes in fish for Validation of Ecological Risk (RECOVER).

164 Impacts of Oil Exposure on Mahi Embryos C. Pasparakis, Rosenstiel School of Marine Sciences / Marine Biology and Fisheries; L.E. Sweet, Environmental Protection Agency USA; E.M. Mager, University of North Texas / Department of Biological Sciences; J.D. Stiegelitz, RSMAS University of Miami / Marine Biology and Ecology; D.D. Benetti, RSMAS University of Miami / Department of Marine Ecosystems and Society; A.P. Roberts, University of North Texas / Department of Biology Institute of Applied Science; J.D. Stiegelitz, RSMAS University of Miami / Marine Biology and Ecology; D.D. Benetti, RSMAS University of Miami / Marine Biology and Ecology; D.A. Crossley, University of North Texas / Department of Biological Sciences; G.K. Cox, J.D. Stieglitz, RSMAS University of Miami / Marine Biology and Ecology; D.A. Crossley, University of North Texas / Department of Biological Sciences; C. Pasparakis

The Deepwater Horizon spill coincided with the spawning window of many ecologically and economically important fish species, such as mahi-mahi (*Coryphaena hippurus*). Aside from the acute mortality elicited by this event, additional sublethal effects may have imparted more subtle yet ecologically significant consequences on populations of pelagic fishes as a whole. The maintenance of embryo buoyancy is crucial to survival, and aids in promoting dispersal by facilitating drift through ocean currents and positioning newly hatched larvae in the upper water columns where planktonic food is plentiful. We found that co-exposure to oil and additional environmentally relevant stressors, such as high temperature and UV-radiation, affect the timing and duration of negative buoyancy in mahi-mahi embryos. Furthermore, impaired negative buoyancy was coupled with significantly faster sinking rates and increased energy depletion, likely resulting in detrimental consequences for these developing fish. The mechanisms behind untimely buoyancy change are unknown, but our findings suggest a behavioral avoidance response as well as an inability to maintain buoyancy due to diminished energy reserves. This research was made possible by a grant from The Gulf of Mexico Research Initiative. Grant No: SA-1520; Name: Relationship of Effects of Cardiac Outcomes in fish for Validation of Ecological Risk (RECOVER).

165 Crude oil impairs heart cell function in the pelagic mahi-mahi (*Coryphaena hippurus*) R. M. Heuer, University of Miami / Marine Biology and Ecology; H.A. Shiels, G.L. Galli, University of Manchester / Faculty of Biology, Medicine and Health Sciences; G.K. Cox, J.D. Stiegelitz, RSMAS University of Miami / Marine Biology and Ecology; D.D. Benetti, RSMAS University of Miami / Department of Marine Ecosystems and Society; M. Grosell, RSMAS University of Miami / Marine Biology and Ecology; D.A. Crossley, University of North Texas / Department of Biological Sciences

Crude oil from the Deepwater Horizon spill of 2010 has been shown to have a number of cardioxic effects across life stages, species, and levels of organization in marine fish. Over the last decade, the use of the mahi-mahi (*Coryphaena hippurus*) to study these cardiovascular impairments has been particularly important, since this pelagic species is both ecologically and economically important, since this pelagic species is both ecologically and economically important in the Gulf of Mexico. Mahi exposed to environmentally relevant crude oil concentrations have shown compromised intact animal performance, including reductions to maximal swimming speed and maximal metabolic rate. In addition, in-situ studies have revealed a ~40% reduction in cardiac output following oil exposure in mahi. Although cardioxic effects have been widely reported, the mechanisms underlying these impairments remain unknown. In the present study, we examined the impacts of crude oil on isolated mahi heart cells to better understand these mechanisms. Contractility of mahi ventricular heart cells was measured via sarcomere shortening using an IonOptix cell recording system. The first objective was to examine cardiomyocyte contractility over a range of crude oil exposures. The second objective was to examine the impacts of crude oil contractility over a range of stimulation frequencies representative of heart rates observed in mahi (~100–180 beats per minute). Exposure to crude oil was found to significantly reduce heart cell contractile function, but was not found to be dose-dependent in the tested range of concentrations (3.0, 6.4, and 12.9 ug l−1 ∑50 PAH). Exposure to crude oil was also found to impair contractility over a range of stimulation frequencies (1.5, 2.0, 2.5, 2.5, 3.0 Hz; 3.6 ug l−1 ∑50 PAH). In addition to contractility, other mechanical aspects of cell contracture function were also examined. Effects to assess the role of circulating catecholamines (adrenaline) as a potential protective mechanism against these impairments is currently ongoing and will also be presented. This research was made possible by a grant from The Gulf of Mexico Research Initiative.
166 microRNA and messenger RNA networks in early life stages of pelagic and nearshore fish species exposed to Deepwater Horizon oil

D. Schlenk, University of California-Riverside / Department of Environmental Sciences and Engineering; G. Xu, UC Riverside / Department of Environmental Sciences - Developmental cardiotoxicity is a complex phenotype observed in a number of fish species following exposure Polyaromatic hydrocarbon (PAH) or oil. While many PAHs elicit cardiotoxicity through activation of the aryl hydrocarbon receptor (AhR). Additional pathways of toxicity have been observed including downregulation of genes that regulate potassium and calcium channels in embryonic heart and high rate of apoptosis. While functional inhibition the channels has been observed following exposure to oil and non-AhR activating PAHs, mechanisms associated with downregulation has not been observed. MicroRNAs (miRNA) play key roles in a number of diverse biological processes including heart development in vertebrates. To test the hypothesis whether miRNA changes may regulate ion channel genes, embryos and larvae of mahi-mahi (Coryphaena hippurus) were treated with High Energy Water Accommodated Fractions (HEWAF) made from source and weathered DWH oil. miRNAs and mRNAs were sequenced from the same pooled animals and expression comparing using advanced bioinformatics with subsequent target organ predictions based on their expression pattern. To examine GO analysis on the target miRNAs was consistent with pathway analysis of miRNAs, predicting disruption of cardiovascular system development after oil exposure and showed that specific miRNA-mRNA interactions may contribute to these effects (Figure 1). Oil caused an overexpression of miR-133a, miR-34, and miR-15b (Figure 2. Enhanced expression of miR-133a correlated to the decrease in the expression of KCNH2 miRNA, which controls the potassium ion transporter that has been observed to be reduced in the cardiac phenotype in multiple fish species following oil treatment. In addition miR-34 and 15b were also upregulated and informative analyses with miRNAs were consistent impairment of eye development (Figure 2). Ongoing analyses of dose response treatments at each hatch larval stages (48 hpf) will provide additional data that will enhance our knowledge of the impacts of oil on fish development. This research was made possible by a grant from The Gulf of Mexico Research Initiative. Grant No: SA-1529; Name: Relationship of Effects of Cardiac Outcomes in fish for Validation of Ecological Risk (RECOVER).

Alternative Approaches to Animal Testing for Ecotoxicity Assessments

167 Early life stages of a vertebrate species as an alternative model for the study of stressors in marine environment

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Early life stages of marine vertebrates have been scarcely used in ecotoxicity testing. The Senegalese sole (Solea senegalensis Kaup, 1858) is a common vertebrate occurring in Eastern Atlantic coastal areas. Eggs of this marine vertebrate can be obtained from aquaculture rearing facilities and used in laboratory as testing organisms. At the end of the first month of life this species completes a metamorphosis, changing from bilateral to flatten shape morphology. Early life stages of aquatic vertebrates are windows of development considered highly sensitive to anthropogenic contamination, including in marine environment. Organic compounds, such as pesticides and personal care products have been increasingly used and directly released to the aquatic ecosystems or indirectly released from wastewater treatment plants. Besides, human activities have been increasingly changing abiotic conditions, such as ultraviolet (UV) radiation. Therefore, in this work we aim to understand the effects of different stressors to early life stages of S. senegalensis, namely of UV radiation and of the organic compounds -4MB, Carbendazim, Linuron and Triclosan, which have potential estrogenic/androgenic and lethal stages of development. While reduction of growth, behaviour and biochemical markers were evaluated as endpoints in two periods of exposure to stressors: a first initial period between egg stage and 96 hpf and a second period during the nearly 15-day full metamorphosis progression of S. senegalensis. Exposure to UV radiation and to the four organic compounds (compounds -4MB, Carbendazim, Linuron and Triclosan) was performed. Our results strongly suggest that S. senegalensis is a promising vertebrate model for early life stage testing, as it requires the evaluation of effects at different development stages. Initial egg stages globally display a higher sensitivity to stressors, presenting lower LC50 and EC50 values. Besides, biochemical markers (cholinesterases and oxidative stress) were differentially affected, depending on S. senegalensis life stage. Significant alterations of normal behavioural pattern were observed in response to stressors exposure, confirming behavior as a sensitive and relevant tool in ecotoxicology studies. The increasing environmental levels of the contaminants tested may lead to adverse effects on highly sensitive life stages of marine vertebrate species.

168 Predicting in vivo toxicity from in vitro transcriptional responses following chemical exposure

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Alternatives to in vivo animal testing in ecotoxicology aim to increase the throughput of chemical safety assessment whilst reducing the number of animals used. The use of in vitro systems is more cost-effective, practical and expedient. However, it is still unclear whether in vitro methods provide the level of information gained from the use of a whole-living system. In vitro to in vivo extrapolation relies on measuring the effects of chemicals on cultured cells or biological molecules to predict how exposure to those compounds might cause adverse effect in animals or people. In this study, we investigated whether the transcriptional state of a trout gill cell line (Oncorhynchus mykiss, RTgill-w1) exposed to a given chemical can be used as a biosensor to predict toxicity in a zebrafish embryo (Danio rerio). More specifically, we developed a regression model linking gene signatures that are independent of compound lipophilicity and predictive of toxicity. We show the ability of residual analysis to identify excess toxicity and to accurately predict in vivo toxicity for most of the chemical MoA in the panel. Our results support the view that gill cell line has the potential to replace zebrafish embryo in toxicity testing.

169 Combining computational modelling with in-vitro cellular responses in order to predict chemical impact on fish growth

K. Schirmer, Eawag / Environmental Toxicology; H. Mottaz, R. Schoenenberger, Eawag Swiss Federal Institute of Aquatic Science and Technology / Environmental Toxicology; J. Stadnicka-Michalak, EPFL Swiss Federal Institute of Technology / Environmental Toxicology

A paradigm shift in chemical risk assessment is needed due to the time-consumption and ethical controversies of current chemical testing on animals. Therefore, in this project, we propose that the chemical effects on cell population growth, measured over few days, can be used as proxy for chemical impact on fish growth, which needs weeks to occur. In particular, we linked information on the proliferation and survival of a fish gill cell line (in vitro) to the effect of chemicals on fish growth (in vivo). Research was divided into two phases. In the first phase, we have tested in vitro several chemical concentrations that correspond to those used in in-vivo experiments. In the second phase an approach was developed in which no prior knowledge about chemical concentrations tested in vivo was required: in vivo data were needed only to validate the model but not to decide which chemical concentrations should be tested in vitro. More than ten organic chemicals (including fungicides, herbicides, insecticides, industrial compounds and pharmaceuticals) were tested for different fish species (rainbow trout, fathead minnow and zebrafish). The results indicate a very good agreement between measurements and predictions determined for different species of fish, being exposed in vivo from 7 to 62 days, depending on the species and test design. Results moreover confirm that it is possible to predict chemical impact on fish growth without prior knowledge on concentrations that are used in in vivo studies for chemicals that do cause an effect on fish weight as well as for those that do not. Therefore, in spite of severe assumptions and simplifications, combining in vitro experiments with computational modelling can result in a powerful strategy for screening chemicals to determine their effects on fish. In addition, considering the simplicity, rapidity and low costs of this approach, we believe that it can be an encouraging step toward alternatives to long-term whole organism toxicity testing.

170 Ecological Threshold for Toxicological Concern (eco-TTC) - Applications for Environmental Risk Assessment in Various Contexts

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wealth of ecotoxicological information as Predicted No-Observed Effect Concentrations (PNECs) on diverse chemical substances in the form of probability distributions. These enable the prediction of untested chemicals based on a structural attribute, mode of action, or functional use. The approach may be useful for assessing chemicals at early tiers of the risk assessment process, providing hazard perspective on chemicals that lack QSARs, guiding product development discussions, and assisting read across or category justifications. An ecotoxicological database of approximately 120,000 records was developed based on recent assessments of published data and international chemical management programs. This ecotoxicity data is associated with physical chemistry data and curated taxonomic information for the organisms tested, including a process to conclude acute and chronic effects as well as identify the PNEC for exposed ecosystems based on depth and breadth of data. Several mode of action schemes are also included to facilitate development of a best approach for grouping compounds. To make these data accessible and useful to stakeholders, the database was transitioned from Microsoft Excel and Access into a modern MySQL format, allowing for a format that is relational and scalable, facilitating easy access, sharing, and integration with other datasets and tools. The database is accessed via a web-based query system that is integrated with PNEC calculator and probability distribution tools. The novel interface allows users to explore the data, upload additional datasets, derive threshold values based on specific criteria, and explore the potential use and application of the ecoTTC concept. An international workshop was held to discuss and evaluate the feasibility of the eco-TTC approach, which included evaluation of several case-studies based on particular decision-contexts (e.g., pre-registration prioritization, chemical risk MoAs, integrated specific risk assessment, mixtures, product development, criteria development). This presentation will highlight the discussions and conclusions from a recent multi-stakeholder workshop, including exploration of how this approach could be applied and integrated into evaluation strategies.

171 Mode of action diagnosis by normalized multiple endpoint assessment in zebrafish embryos
E. Teixido, Helmholtz Center for Environmental Research - UFZ GmbH / Department Bioanalytical Ecotoxicology; N. Klüver, Helmholtz center for environmental research - UFZ / Department of Cell Toxicology; O. Kerkhof, Helmholtz centre for environmental research - UFZ / Department Bioanalytical Ecotoxicology; M. Leonard, IOREAL SA; T. Kießling, Scientific Software Solutions; R. Altenburger, UFC Centre for Environmental Research / Department Bioanalytical Ecotoxicology; S. Scholz, Helmholtz Centre for Environmental Research / Department Bioanalytical Ecotoxicology

The diagnosis of a specific or reactive mode of action (MoA) is crucial for the prediction of adverse effects using alternative test systems. While baseline toxicity can be easily predicted using regression-based QSARs, the identification of non-baseline toxicants requires assessment of endpoints that can be related to the MoA of a chemical and ultimately assign an adverse outcome pathway. Such a MoA analysis could be possible using the zebrafish embryo test (ZFET) extended by various endpoints covering chemical risk MoAs. In the specific risk assessment system, the detection of MoA-related endpoints has been discussed to improve its predictive capacity for acute and chronic fish toxicity, and for human developmental toxicity. We hypothesized that using a battery of endpoints in the zebrafish embryo test would allow to differentiate between baseline toxicity, formation of methemoglobin, neurotoxicity, heart rate inhibition, and developmental toxicity. Therefore, we compared the toxic ratios and endpoint-specific effect concentrations (EC50) of 12 compounds representing 5 broad MoA groups with 2, respectively 4 (neurotoxicity) compounds per MoA. In order to compensate for differences in the toxicokinetics and mortality, the effect concentrations were normalized by the LC50 of each compound. It was shown that the toxic ratio and effect concentrations for behavior, heart rate inhibition and chorda malformations were able to differentiate the selected compounds according to their anticipated MoA. Using a threshold for the normalized effect concentration a decision tree was developed that allowed to assign a MoA to a compound. A major bias of the selected approach could be the variability associated with visual phenotype assessment – which may depend on the experience and accuracy of the observer. Therefore, we developed a software named FishInspector that enables a more unbiased assessment of malformation using images of zebrafish embryos. Using the software and a system for automated positioning of zebrafish embryos we compared the phenotypes of 25 compounds with known developmental toxicity outcome in rats and/or rabbits. In contrast to the previous analysis, we applied a different normalization approach based on the most sensitive endpoints. The analysis indicated that the developmental toxicity observed for inhibitors of cyclooxygenase may not be related to the pharmacological MoA; given the diverse phenotype patterns observed for this class of compounds.

172 Poster spotlight: MO158, MO159, MO190

Migratory bird species at risk - the role of pesticides and other chemicals

173 CMS talk setting the scene for the CMS working group on poisoning and outlining CMS needs in terms of scientific input from SETAC
B. Heredia, UNEP/CMS / Avian Unit

174 Main scientific gaps in knowledge of risk from pesticides to [migratory] wildlife globally, and potential contribution of WTIG to CMS questions - POSTER SPOTLIGHT MO456
C.A. Bishop, Environment and Climate Change Canada / Wildlife Research Division

175 Main scientific gaps in knowledge of risk from rodenticides to [migratory] wildlife globally, and potential contribution of WTIG to CMS questions - POSTER SPOTLIGHT MO457
P. Berny, VETAGRO-SUP / Toxicology

176 Main scientific gaps in knowledge of risk from Pb ammunition and shot to [migratory] wildlife globally, and potential contribution of WTIG to CMS questions - POSTER SPOTLIGHT MO458
R. Croninig, Wildfowl & Wetlands Trust

177 Main scientific gaps on knowledge of NSAIDs [migratory] wildlife globally, and potential contribution of WTIG to CMS questions - POSTER SPOTLIGHT MO459
M. Taggart, University of the Highlands and Islands / Environmental Research Institute

178 Main scientific gaps on knowledge of deliberate poisoning to [migratory] wildlife globally - POSTER SPOTLIGHT MO460
M. Odino, Independent Environmental Services Professional

179 Questions and discussion

180 Regulatory view describing the extent to which [if any] regulation takes into account neighbouring country/regional use of compounds, accounts for how local use might affect migratory species, how field data on migratory species might feed into regulatory
R. Sharp, EFSA - European Food Safety Authority / Pesticides Unit

181 Panel discussion with audience and presenters focusing on how SETAC can interact with CMS usefully to provide scientific evidence and expertise

Challenges in setting, meeting and measuring specific protection goals for plant protection products

182 Towards a more holistic environmental risk assessment approach of crop protection products as tools in agriculture
P. Dobmen, BASF SE / Environmental and Consumer Safety, Ecotoxicology

Currently, very complex risk assessment approaches are conducted for crop protection products. This is largely justified as pesticides are intentionally introduced into ecosystems and since these substances are intended to have effects on “crop pests” and side effects on non-target species cannot be fully avoided. However, doing so, we often ignore the context, the reasons why crop protection products are utilized. This ignorance can lead to failing the overall targets or lead to sub-optimal or even wrong decisions. This will be explained in more detail in the following presentation advocating for a more holistic approach in environmental risk assessment. Agriculture per se does have an impact on the environment and
does reduce biodiversity at the sites used for food production. This, in principle, is true for any kind of agriculture. As a consequence, it is irrational to demand that agriculture, respectively the different agricultural practices, shall have ‘no impact’ on the environment. Accordingly, regulations require that ‘no unacceptable’ impact may occur. To define what constitutes an acceptable impact and what not, the ‘Ecosystem Services’ concept is considered a suitable approach. This is not a call for ignoring the impact of crop protection products. However, for achieving our targets and maintaining or even increasing the sufficient local food production and an environmental impact we need to have a broader approach and evaluate the cost/benefits of all agricultural tools equally against the background of food produced per area. Scientific tools to go for such more complex holistic approaches are partly available, but more efforts are needed to develop practical and manageable concepts allowing an avoidance of chemicals in large areas. In addition, the regulatory options have to be broadened from mere risk assessment of chemicals towards a landscape level assessment of food production, and risk management has to include landscape management options. Ecotoxicological research is undoubtedly important; however, in order to achieve the overall target of feeding human populations in a sustainable way and maintaining ecosystem health, we need to cover all aspects of ‘stress ecology’ (impacts of humans on ecosystems) beyond the impact of chemicals. Therefore this call for a more holistic environmental risk assessment approach.

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Identifying ecosystem services-based protection goals.
L. Malby, Y. Pan, The University of Sheffield / Dpt of Animal Plant Sciences
There is an increasing interest in the use of ecosystem service-based approaches for assessing the risk of environmental contaminants to ecological systems. Ecosystem functions become ecosystem services when they are utilized and valued by people. Therefore, the first step in implementing an ecosystem services approach to ecological risk assessment is to identify what portfolio of services are required, by whom and where they should be protected. But what preferences should contribute to identifying the services to be protected and how should preferences be assessed? These questions are particularly pertinent when considering the role of the general public in protection goal prioritization. Here we use information from case studies in the UK and China to investigate the importance of: who you ask (do different sections of society have different preferences?); what they know (how does prior knowledge influence preferences?); how preferences are assessed (e.g. stated v revealed preferences). We demonstrate that all three factors can have an impact on which freshwater ecosystem services are preferred by the general public and therefore should be prioritized for protection.

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ECPA over-arching Specific Protection Goals proposal for EFSA Non-Target Terrestrial Plants, Non-Target Arthropods and Soil Organisms Guidance Documents based on EFSA Ecosystem Services approach
K. Romijn, Bayer CropScience AG
Currently there are 3 Scientific Opinions from EFSA that are waiting to be developed into Guidance Documents i.e Effects of pesticides on Non Target Terrestrial Plants, Non Target Arthropods and Soil Organisms. Whilst each of these Scientific Opinions makes proposals for SPGs, the European Commission and Member States should agree on the on SPGs before they can be taken forward to be used in the Guidance Document development phase. The purpose of this paper is to provide industry input for consideration and discussion during this process. In an earlier EFSA Scientific Opinion it was recommended by EFSA that Specific Protection Goals (SPGs) should be based on the principle of Ecosystem Services utilising 6 dimensions: (i) biological entity, attribute, magnitude, temporal and geographical scale of the effect, and the degree of certainty that the specified level of effect will not be exceeded. Whilst this EFSA Scientific Opinion is a good basis for setting SPGs going forward, the experience with the EFSA Bee Guidance Document shows there is a need to reconsider how the principles described in this EFSA SPG Opinion are applied to SPG setting in individual Guidance Documents. In the case of the EFSA Bee Guidance SPG it was not the definition of “negligible effects” on colony strength that was the controversial issue but the translation of this into a numerical value (< 7%) without robust scientific justification. The use of 7% was suggested there was data to support it but in fact it was still a judgement, i.e. it is a hidden ‘judgement’. The suggestion that there is a ‘linear’ or arithmetic relationship between large (>35%), medium (15-15%) small (7-15%) and negligible (3.5-7.5%) is in itself surprising for a biological system particularly one with complex feedback loops such as in honeybee colonies. To avoid the difficulty of selecting a specific hard numerical SPG value upon which consensus between different scientist and stakeholders is based it is recommended to have an explicit qualitative judgement adapting the EFSA Ecosystem Services approach. The predicted impact of any effect of a PPP on an invertebrate/plant population should be described using expert judgement, which combines the predictions of all 4 EFSA dimensions above (attribute/nature of effect, magnitude, temporal and spatial scales of effects) as well as the number and importance (eg. stakeholder species) of species potentially affected, and the frequency of occurrence.

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Protection goals for non-target terrestrial plants: Is in-field protection of beneficial weeds achievable?
J. Davies, Syngenta / Environmental Safety; L. May, A. Russell, A. Seville, D. Stock, Syngenta
EFSA’s Scientific Opinion addressing the state of the science on the risk assessment of plant protection products for non-target terrestrial plants (NTPTPs) was published in 2014. The Opinion defines non-target plants as all plants growing outside fields and those growing within fields that are not the intended pesticide target. It also states that non-crop plants growing in in-field areas provide ecosystem services including food web support, aesthetic value, genetic resources and endangered species, which require protection from the adverse effects of plant protection products. As such, The Opinion advocates the protection of plant species growing in-field that under current agricultural practice would be considered target weeds, as well as those growing in the crop canopy. In addition, the regulatory options have to be broadened from mere risk assessment of chemicals towards a landscape level assessment of food production, and risk management has to include landscape management options. Ecotoxicological research is undoubtedly important; however, in order to achieve the overall target of feeding human populations in a sustainable way and maintaining ecosystem health, we need to cover all aspects of ‘stress ecology’ (impacts of humans on ecosystems) beyond the impact of chemicals. Therefore this call for a more holistic environmental risk assessment approach.

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Specific Protection Goals and the Assessment of Key Drivers in the Aquatic Environment: Are we doing the right thing?
R. Benstead, Fera Science Ltd. / Centre for Chemical Safety and Stewardship; D. Phillips, Fera Science Ltd / Environmental Sciences; P. Gilbertson, Fera Science Ltd; J. Clunn, Centre for Crop Health and Protection (CHAP)
When designing High Tier Assessments, the underlying concept is a progression from simple and conservative laboratory exposures, towards those that more closely resemble the ‘Final Reference’ (the actual ecosystem), so that the risk assessment can be refined by reducing the Assessment Factor that accounts for uncertainty. In the aquatic environment, the focus of High Tier Assessments should not be on aquatic Protection Goal (AG); rather it is defined for surface water quality targets and Ecosystem Services. Biodiversity is a common and important General Protection Goal, and Ecosystem Services, as they pertain to human health and welfare, are comprised of ‘provisioning services’ (e.g. drinking water), ‘regulating services’ (e.g. water purifying microbial communities), ‘supporting services’ (e.g. organisms contributing to nutrient cycling) and ‘cultural services’ (aspects of the water landscape that conserve species and promote well-being). To this aim, this presentation will illustrate that the feasibility of protecting weeds in-field requires investigation of complex species definitions and agri-cultural consequences while the discovery and availability of new herbicides with the required selectivity is considered unlikely, particularly under the current regulatory framework in the EU.

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Is “biodiversity” a measurable study endpoint?
E.M. Bakker, Eurofins-Ictina
The general protection goal addressed by current and future risk assessment schemes for Plant Protection Products is Biodiversity. In addition to this there are specific protection goals that may conflict with the general protection goal. Specific protection goals are generally phrased in terms of Ecosystem Services and expressed in human currency there is a risk that economic motives may prevail in their prioritization. After all, who is to judge e.g. whether preservation of pest control capacity outweighs the preservation of aesthetic value? Biodiversity is a concept with many facets and the quantification of biodiversity is no straightforward exercise. A vast array of indices exists (see e.g. Maguran 2004)

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and not a single one would capture the essence on its own. The two basic parameters involved in biodiversity measures are the number of species and their abundance. Their correct estimation is core to biodiversity assessments and their balance describes community structure in terms of e.g. dominance, evenness and diversity. Similarity indices have been developed that enable the comparison of (sub)habitats, which seems a promising lead to assessing effects on biodiversity in experimental designs. With this contribution we explore different approaches to quantifying effects on structure and function caused by artificial protection products in an experimental setting. We apply and compare multivariate statistical approaches, similarity indices and a combination of univariate statistics and species richness assessments and discuss how these findings may address the general issue of effects on biodiversity.

Innovative techniques for enhancing and monitoring microbial activities for in situ remediation of contaminated sites

188 Evaluation of plant-driven biostimulation of soil microbiota for the setup of a site-tailored rhizoremediation process in a historical PCB-polluted soil L. Vergani, University of Milan / DeFENS; F. Mapelli, University of Milan-DeFENS / Department of Food, Environmental and Nutritional Sciences; E. Terzaghi, University of Insubria (Como) / Department of Science and High Technology, Como; G. Raspa, Sapienza University of Rome / Department of Chemical Engineering Materials and Environment; O. Uhlík, University of Chemical Technology Prague; E. Zanardini, C. Morosini, University of Insubria / DSAT; A. Di Guardo, University of Insubria / Department of Science and High Technology; S. Borin, University of Milan / DeFENS

The Site of National Priority (SN) Brescia-Caffaro is a highly polluted area in Northern Italy presenting mixed and uneven soil contamination by metals and organic pollutants, in particular polychlorinated biphenyls (PCBs). In order to evaluate the biostimulation potential of plant species and to evaluate possible treatments for the development of a suitable rhizoremediation strategy, an experimental trial including ten vegetated treatments and their non-plant cultivated controls was set up for 18 months in greenhouse conditions. Molecular fingerprinting was applied to unveil the ability of different plants/soil treatments to shape the structure of soil microbial communities. The results showed a succession over time in both bacterial and fungal assemblages. Only the diversity of the bacterial community was, nevertheless, significantly and differentially influenced according to the applied treatment. The stimulation effect on the organic matter hydrolytic activity of the soil microbiota was evaluated using fluorescein diacetate as a proxy. All the vegetated treatments showed a significant increase in activity after 18 months from planting, demonstrating effective biostimulation of the soil bacterial communities, putatively enhancing their degradation capacity and, consequently, sustaining rhizoremediation. Aiming to select bacterial strains to be exploited for autochthonous bioaugmentation coupled to rhizoremediation, we established a collection of isolates from the soil biostimulated by Phylaitis arundinacea. This species cultivated in conditions of redox cycle showed to stimulate the highest increase in soil bacterial biomass after 3 months from planting. Moreover, with the 18-month biostimulated soil was incubated with 13C-labelled 4-chlorobiphenyl, the production of 13CO2 indicated metabolic activity of biphenyl and possibly the presence of PCB-degrading populations. All the isolates were identified as Actinobacteria and were characterized for PCB-degradation and plant growth promotion. In particular, three Rhodococcus sp. strains significantly promoted lateral root development in the model plant Arabidopsis thaliana and depleted PCBs from the cultivation medium according to the results of a resting-cell assay, thus representing ideal candidates to sustain PCB-rhizoremediation through a site-tailored bioaugmentation approach.

189 Enhancement of Biological Reductive Dechlorination by in situ Adsorption onto Colloidal Activated Carbon: from the Lab to the Full Scale Application M.P. Papini, F. Arjmand, Università La Sapienza / Chemistry; P. Ciampi, C. Esposito, Università La Sapienza / Department of Earth Sciences; M. Carboni, P. Goria, J. Bursting, Regenesi Ltd; S. Rossetti, B. Matturro, Water Research Institute Italian National Research Council IRSACNR; M. Bacchi, P. Foglio, Rain Ferroviria Italiana - Technical Laboratory (RFI)
The University of Rome “La Sapienza” has been commissioned to evaluate strategies for the management of the contaminated areas of the new High Speed Railway Station of Bologna (Italy), where a historical Chlorinated Aliphatic Hydrocarbons (CAHs) contamination has been found in two aquifers and characterized by a long-term contamination pattern (PCBs, TCE and cis- and trans-DCE - concentrations ranging between 10-100 µg/L). The Italian environmental legislation is among the most restrictive in Europe with some of the most stringent target levels especially concerning the CAHs. A thorough investigation of the site has been carried out (geological, chemical and biological) and integrated with a microcosm study. Based on the results, biological reductive dechlorination was recognized as a potential approach for the site remediation but the extremely low CAHs concentration and the consequent kinetic limitation made it unfeasible for the site. Thus, the possibility to use a new dispersed colloidal activated carbon technology (Plumestop®), Regenesi together with an electron donor to create an in-situ adsorption/biologically-reactive zone was deeply investigated and finally adopted as the site-specific remediation approach. The full-scale remediation plan was approved by the local authorities and completed by the end of 2016. This was the first example of a completed full-scale application of this approach in Europe and the monitoring results after more than one year appear particularly encouraging. A strong reduction of CAHs concentration and a rapid decrease of carbon mass were observed in all the treated zones. Together with classical chemical analyses, microbiological tools, such as qPCR and CARD-FISH, were used to verify the enhancement of the biological reductive activities induced by the simultaneous injection of activated carbon and electron donor.

190 An innovative bioelectrochemical reactor for in-situ treatment of groundwater contaminated by monoaromatic petroleum hydrocarbons E. PALMA, CNR-IRSA; M. Daghio, A. Franzetti, University of Milano - Bicocca / Department of Earth and Environmental Sciences; M.P. Papini, Università La Sapienza / Chemistry; F. Aulenta, National Research Council / Water Research Institute (IRSァ)

A major share of world energy production, derives from fossil fuels, such as oil. According to OPEC (Organization of the Petroleum Exporting Countries), world oil demand growth is expected to rise by 1.53 mbd in 2017. The large-scale use and countless applications of petroleum compounds, frequently lead to environmental contamination, as a consequence of petroleum transport, storage and refining, or accidents. Groundwater contamination by petroleum hydrocarbon is a serious problem, with nearly 50% of groundwater contamination being due to petroleum-derived products such as mineral oil, chlorinated hydrocarbons, monoaromatics (e.g., BTEX) and polycyclic aromatic hydrocarbons (i.e., PAH). Accidental petroleum spills may result in severe environmental problems, hence reducing the development and implementation of suitable remediation strategies. In recent years, microbial electrochemical technologies (MET) have attracted considerable attention as an effective and sustainable remediation technique. In MET the microorganisms catalyze oxidation or reduction reactions by using solid-state electrodes as terminal electron acceptors or donors. The discovery that carbon-based electrodes can be used as terminal electron acceptors for the anaerobic oxidation of a variety of organic substrates has raised the possibility that they could be employed in-situ to accelerate the anaerobic oxidation of environmental contaminants, such as petroleum hydrocarbons in soils and groundwater. Here we describe a novel bioelectrochemical reactor configuration, named the “bioelectrochemical well”, that is suitable for in-situ treatment of petroleum contaminated groundwater. A lab-scale prototype of the bioremediation cell ("bioelectrochemical well [1]") has been realized and operated in a continuous-flow regime using first toluene and then a mixture of BTEX as model contaminants. The performance of the bioelectrochemical reactor was characterized in terms of degradation rate and yield. GC-MS analysis was also carried out in order to shed light on the "electrogenic" pathway of contaminants biodegradation. This study was financially supported by Fondazione Cariplo in the framework of the project BE-ERAGE - BioElecTrochemical Remediation of Groundwater plumes (2015-0195). [1] Palma E., Daghio M., Franzetti A., Petrangeli Papini M., Aulenta F. The bioelectric well: a novel approach for in situ treatment of petroleum-contaminated groundwater. Microb Biotechnol., 2017. doi: 10.1111/1751-7915.12760.

191 Identification of major HMW-PAH degrading communities during active bioremediation of a PAH-contaminated soil J. Vila, Instituto de Recursos Naturales y Agrobiología; M. Grifoll, Universitat de Barcelona / Dept. Genètica, Microbiologia i Estadística; M. Aitken, University of North Carolina / Environmental Sciences and Engineering; J. Ortega-Calvo, Instituto de Recursos Naturales y Agrobiología / Agroquimica y Conservacion del Suelo

Biodegradation of polycyclic aromatic hydrocarbons (PAHs) in soils is generally constrained by their low availability to microbial communities. As a result, a range of unpolluted conditions of PAHs in soils were created, with a major composition in high molecular weight (HMW) compounds (four or more rings). We analyzed the active microbial processes associated with the dissipation of PAHs during a lab-scale bioremediation of a creosote-contaminated soil. Treatment under stimulated conditions resulted in an extensive reduction (93%) of the total PAH concentration. Low molecular weight (LMW) compounds (2 and 3-rings) were mostly depleted during the first month of incubation. In contrast, degradation of HMW-PAHs started thereafter following biphasic kinetics. Despite the 16S rRNA gene copy numbers (bacterial abundance) remained constant throughout the 5-month treatment period, the number of 16S rRNA gene transcripts (bacterial activity) dramatically increased (from 10^7 to 10^10 copies/g dry soil) during the initial 45 days, associated with major PAH removal. 16S rRNA gene pyrosequencing revealed distinctive signatures for and among communities that evolved with time. Gene expression analysis of ring hydroxylating dioxygenases, together with changes in pyrosequencing libraries, identified members of Pseudomonas as the main LMW-PAH degraders. In contrast, dioxygenases of
Gram-positive bacteria, associated to Mycobacterium, were mainly active during the last two months of incubation, when only residual fractions of HMW compounds were degraded. Community analysis during the period of major HMW-PAH removal identified members of the recently described order Immobilineisolacticibacterales and members of Sphingobium as the main active populations. Their role on HMW-PAH removal was confirmed by DNA-SIP. Members of Immobilineisolacticibacterales were the major phyotypes detected in the acidification process, where members of Immobilineisolacticibacterales clearly predominated in incubations with 13C-pyrene and 13C-benzo[a]anthracene. Interestingly, members of Mycobacterium, traditionally associated to HMW-PAH degradation were not detected in either of the incubations. Our results indicate the minor contribution of Mycobacterium to the degradation of the more labile fraction of HMW-PAHs. Their increase in activity during the late incubation phase, when degradation kinetics were drastically attenuated, suggested their potential action on the residual fraction of contaminant.

192 Stable Isotope Raman Microspectroscopy and Surface-Enhanced Raman Scattering: Analysis of Microorganisms at Single Cell Level
N.P. Iveyka, Technical University of Munich / Chemistry Department, Chair of Analytical and Water Chemistry; R. Weiss, M. Seidel, Technical University of Munich; R. Niessen, Technische Universität München / Chemistry Department, Chair of Analytical and Water Chemistry; M. Elsner, Technical University of Munich / Chemistry Department, Chair of Analytical and Water Chemistry Microbes signal intensity and discrimination in most ecosystems. They are essential for global biogeochemical cycles and for biodegradation of soil and water pollutants. Therefore, it can be crucial to develop reliable and sensitive methods for the detection, discrimination and identification of microorganisms as well as for analysis of their activity. Raman microspectroscopy (RM) as an emerging tools for the nondestructive characterization of the chemical and isotopic composition (due to a red-shift in Raman spectra for heavier stable isotopes) of microorganisms at the single cell level.[1-3] RM allows for in situ investigations of ecophysiology and metabolic functions of microbial communities. Furthermore, the sensitivity of RM and SIRM analysis can be significantly improved (in the range of 105 to 107, max. up to 107) due to surface-enhanced Raman scattering (SERS), e.g. by using of Ag nanoparticles synthesized in situ. In contrast to RM and SIRM (where whole-organism fingerprints for bacteria are obtained), SERS is more selective and provides information on cell surface substances. We applied SIRM and SERS for analysis of unlabeled, 13C- and 15N-labeled single bacterial cells.[4-6] Single cell SIRM analysis was carried out for the Delaglumobacter spec. strain N47, a strictly anaerobic sulfate-reducing, degrading the recalcitrant environmental pollutant naphthalene. For the 13C-labeled N47 cells peak pattern from isotopologues of phenylalanine with 0, 2, 4 and 6 13C atoms was found even though this strain is a strict anaerobe growing on 13C-naphthalene. Additionally, our results suggest an incorporation of hydrogen carbonate from the medium into biomass during growth of strain N47 on naphthalene. Furthermore, SERS analysis of E. coli revealed that the SERS signal intensity depends on different factors (storage time, presence of D-O) and can reflect the metabolic activity of cells. Our findings can open new possibilities for the application of SERS (in combination with a stable isotopic approach) to probe for the activity of pollutant degrading microorganisms at the single cell level. [1] Berry B, et al. 2015. Proc Natl Acad Sci USA 112: E194-E203. [2] Wang Y, et al. 2016. Curr Opin Biotechnol 41: 34-42. [3] Ivanov NP, et al. 2015. J Biol Chem 290: 4335-4375. [4] Kubryk P, et al. 2015. Anal Chem 87: 6622-6630. [5] Kubryk P et al. 2016. Analyst 141: 2874-2878. [6] Weiss R, et al. in prep.

193 Flux chambers data for the estimation of the biodegradation rate in the subsurface at hydrocarbon contaminated sites
I. Verginelli, University of Rome Tor Vergata / Department of Civil Engineering and Computer Science Engineering; R. Pecoraro, Versalis; R. Baciocchi, University of Rome Tor Vergata
The occurrence of aerobic biodegradation in the subsurface by ubiquitous soil microorganisms has been identified and is accepted as the main mechanism for the dissipation of petroleum hydrocarbon vapors from contaminated soil or groundwater on outdoor and indoor air quality. The occurrence of natural attenuation in the subsurface is generally evaluated by employing multi-level soil gas sampling through the installation of nested probes, by applying the so-called “gradient method”. In this way, it is possible to assess the vertical concentration profiles of vapors and oxygen at the source zone and hence to evaluate the attenuation rates in terms of soil gas fluxes through the subsurface. In this work, we propose a novel approach based on the combination of the data obtained from standard source characterization with dynamic flux chambers measurements. The natural attenuation rates are calculated as difference between the flux of contaminants estimated with a non-reactive diffusive model starting from the concentrations of the contaminants detected in the source (soil and/or groundwater) and the effective emission rate of the contaminants measured using dynamic flux chambers installed at ground level. The reliability of this approach was tested in a versus site characterized by the presence of BTX in soil and groundwater, using dynamic flux chambers. The site is characterised by the presence in the subsurface (mainly in groundwater) of BTEX and light petroleum hydrocarbons. The flux of volatile organic compounds (VOCs) from the subsurface was estimated using 14 “dynamic” chambers, by measuring with a canister the concentration of vapours collected over a period of approximately 6 hours. Before starting the measurement, the achievement of steady-state conditions inside the chamber was assured by purging at least 4 chamber volumes with an inert gas. The measurements sampling points were repeated in 4 seasonal campaigns. The obtained results highlighted that the traditional methods based on the application of a non-reactive diffusive model with the concentrations measured in the soil and/or groundwater can lead to an overestimation of the emission rates of BTEX from the subsurface in some cases up to 4 orders of magnitude. Furthermore, the BTEX loss rates for the investigated site were found to be up to almost 0.5 kg/year/m2. These rates are in line with the values reported in the recent literature for natural source zone depletion.

New frontiers in Life Cycle Inventory data collection and modelling

194 The end of an era: is data and model exchange across LCA software tools finally possible?
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In the context of the Environmental Footprint (EF) pilot phase, the European Commission requested the development of 70 representative products/organisations (RPs/ROs) so they are consistent with the requirements of the final product environmental footprint category (PEFCA) and organization environmental footprint sector rules (OEFSRs), they use the prescribed EF-compliant secondary datasets, and can be made available for free to any user applying a PEFCR/OEFSR and are easily usable in the major existing LCA software tools. An implicit prerequisite is that software tools give equal results when calculating the impact of the same RPs/ROs. At the start, a document was established that defined the minimum requirements as well as the verbs for modeling. This way, potential problems for implementability of the models in the different software tools were identified and their use was banned. Five software tools were included, GaBi, openLCA, RangelCA, SimaPro, and Umberto. The next step was to test the EF impact assessment (IA) results of all EF-compliant secondary datasets. This way, differences coming from their implementation or of the EF IA method could also be identified before testing the results of the models. Furthermore, the development of an extension to the International Reference Life Cycle Data System (ILCD) format (eILCD) enabled importing and exporting models between software tools. Finally, the models developed for RPs and ROs will be tested in the five software tools. The problems faced in importing, calculating and comparing the results of the developed models in the different software tools together with suggestions for overcome these was addressed. Some claim different results in different software tools used, often generally pointing to different “software”, without being more specific. The reason may be rather bound to methodology, age, version, flow list and import-export-interface aspects, or even a combination. This work is the basis to enable the reduction of software-system related issues and makes it easier to detect and prevent mistakes. Most importantly, now there is the commitment of the 5 tool developers to make available to all users in the course of 2018 a compliant import and export interface for the eILCD format. We believe this is a crucial step towards the exchange of data and models across software tools and the comparability of results.

195 LCA using real time information: the case of DEA-enabled monitoring of WWTP lifecycle environmental performances
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Life cycle assessment (LCA) is undergoing the effects of a paradigm shift with the introduction of life cycle costing (LCC) and the power of crowd-sourced information. However, how to use this huge amount of data in a consistent way to obtain more precise, spatially and temporally differentiated life cycle inventories (LCIs) and life cycle impact assessment (LCIA) results is still not an easy task. In the case of wastewater treatment, the larger and larger availability of on-line measurements coming from sensors installed in many wastewater treatment plants (WWTPs) should theoretically enable daily benchmarking, ultimately allowing faster correction actions, when needed. However, WWTPs data still finds very limited use and is often simply stored. The aim of this work is to showcase an application of
temporally differentiated LCA performed with real-time high frequency data and present a methodology for the on-line assessment of the shift in the performance of WWTWs. The implemented methodology performs a DEA-based benchmark coupled with LCA to evaluate the environmental impacts linked to the operation of a WWTP. A web application of the system is currently being developed using the Shiny R package. It will enable plant managers to calculate and visualize LCA results in real time by modifying customizable parameters.

196 Enhancing Land Use Change modelling with IO data
J. Schmidt, Aalborg University / Department of Planning; M. De Rosa, BONUS1 / Agroecology
Land Use Changes (LUC) are responsible for around 11% of global GHG emissions, nearly the same as the transport sector. This is about half of the GHG emissions from coal-based electricity production worldwide. Nevertheless, LUC are often excluded from LCA studies because ascribing the LUC to their drivers distinguishing between production sectors is challenging and requires a complex global inventory data modelling. In order to address this, 2–0 LCA consultants have been developing a model for indirect LUC (iLUC) modelling in LCA since 2011 as part of a crowdfunded project. Recently, the model has been integrating into the multiregional hybrid Input-Output model EXIOBASE, thus providing an unprecedented level of detail in iLUC modelling. Differentiation between use of land among regions of the world is based upon information on potential land productivity in different locations. The IO data allow identifying the land supplied by each country (as a mass flow) along with the land use trends. The agricultural and land use module in EXIOBASE make use of FAOSTAT data, which provide time series on area and production per crop. The data allow modelling the global supply of land to the global market for land, distinguishing between land expansion (land transformation) and land intensifications (increased production per unit of land). The land transformation and intensification LCA activities are populated with data on carbon stocks of different land use types in all countries, and time series of fertiliser use in all countries. The current version of the model (version 4.3) includes the following elementary flows: emissions of CO₂, N₂O, NOₓ, NO₃⁻, NH₃ and resource inputs of accelerated denaturisation caused by transformation of land. The iLUC model can be combined with any life cycle impact assessment (LCA) model. Overall, the results show that for agricultural crops, iLUC increases the GHG emissions with 100–200%, for beef cattle 20–60%, for pigs 40–80%, for dairy products 40–60%, for wood products 50–300%, and for primary plastics 2-15%. The model is location agnostic and can be applied to any decision-making context concerning long-term effects of small-scale changes. The iLUC framework is now integrated in the global EXIOBASE model, thus accounting for all crops in all countries in the world. It has already been applied to more than 50 LCA studies and on several product categories.

197 WSMix: a globally regionalised Water Supply mix framework with current and prospective databases for use in LCA
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Freshwater comes from different sources that are unevenly distributed in the world and different water users (e.g. domestic, agriculture, industry) need different water qualities provided by local (surface, groundwater, rain), external (inter-basin transfers) and alternative sources (e.g. sea water). Water from these sources are withdrawn and processed via water technologies using the local electricity mix. The combination of water sources and technologies results in a regional water supply mix (WSMix) for each specific use. Current LCI databases do not include these mixes when modelling processes leading to a poor representation of water supply systems and related environmental impacts in LCA. Furthermore, changes in water sources, caused by changes of climate and socio-economic factors, will occur in the future. These changes should be considered in LCA of products or infrastructure with long lifespans.

The work aims to develop a WSMix framework for modelling current and prospective WSMixes (WSMix and P-WSMix) and an inventory database for direct use in LCA. To demonstrate the relevance of including WSMix and P-WSMix in LCI databases, case studies have been conducted. To develop the WSMix framework, system boundaries have been defined and variables in classification and terminology of water sources and users have been harmonized. A WSMix database for different users has been developed and a technological matrix has been established to link water sources to water production technologies and energy use. To develop the P-WSMix, a methodology based on algorithms enabling to obtain prospective WSMix (P-WSMix) is proposed. Data on water demand and water availability projections for different scenarios and time horizons have been used. The WSMix includes a framework, a WSMix database and technological matrix. The P-WSMix includes also a framework, a P-WSmix database and electricity mix and technology evolutions. The WSMix database covers 93 countries at different spatial scales for various users. The P-WSmix covers 73 countries at national scale for two users under different scenarios and time horizons. It has been shown that the environmental impacts of supply public water are highly dependent on the country and change over time. The inclusion of WSMix and P-WSmix in LCI databases is relevant for a more consistent water-use related impact assessment and for the LCA of infrastructures or products with a long life span.

198 The evolution of database- and tool development for Agri-Footprint
B. Durlinger, L. Kuling, Blonk Consultants
From performing individual Life Cycle Assessment (LCA) studies for specific products, the field is moving towards automated LCAs for full product portfolios and tool and database development. This ongoing evolution is a result of the increased quality and availability of background databases as well as an increased acceptance of LCA as the measurement and monitoring tool for environmental impact. However, a point has been reached where existing LCA software and data structures have become a limiting factor for further development. Therefore, we would like to present our recent developments regarding database and tool development for LCA purposes. Existing LCA software frameworks have become limiting in our database development, because they only have a limited set of calculation features and interfacing capabilities. Also, the data structure of existing LCA software has proven to be limiting. For example, there is no explicit distinction between a process, products/substances, and exchanges. This can result in loss of valuable information. Therefore, we have decided to develop our own database infrastructure and accompanying calculation and import/export modules, that provide enhanced flexibility. This allows for more freedom, we can now make our own choices on how data is stored, what types of analyses can be performed and how this information is presented to a user. In addition, we see a trend where LCA analysts are becoming more and more interested in advanced tools that utilise Life Cycle principles. For Agri-Footprint 2018 we are therefore developing a completely new framework in a Python/Django environment that aims leverage the past developments of Blonk Consultants and Agri-Footprint and utilise them to develop a cloud based Life Cycle Inventory datastore and calculation engine to support and improve both our internal data developments and to serve as a backbone for custom tools for users. With this presentation we hope to contribute to the advancement of LCA databases and tools by providing insight in recent Agri-footprint developments.

199 Poster spotlight: TU097, TU098
Behavioural Ecotoxicology: Unravelling behavioural responses to chemical contaminants in the environment
E. McCallum, Umea University; A. Sundelin, J. Fick, Umea University / Department of Chemistry; A. Alarntä, Swedish University of Agricultural Sciences SLU / Department of Wildlife, Fish & Environmental Studies; G. Hellström, Swedish University of Agricultural Sciences SLU; T. Brodin, Umea University / Department of Ecology and Environmental Science
Concern over the impact that pharmaceuticals have on wild aquatic organisms has increased over the past decade. Laboratory studies have shown that pharmaceuticals can cause sub-lethal changes to animal behaviour and physiology; however, few studies have addressed whether effects documented in the laboratory extend to the natural environment. We exposed fish to one of two pharmaceuticals (temazepam and ibersartan) commonly detected in the environment at two doses (80 ng/L, 1500 ng/L) - 200%, for dairy products 40% for wood products 50-300%, and for primary plastics 2-15%. The model is location agnostic and can be applied to any decision-making context concerning long-term effects of small-scale changes. The iLUC framework is now integrated in the global EXIOBASE model, thus accounting for all crops in all countries in the world. It has already been applied to more than 50 LCA studies and on several product categories.

200 Do laboratory assays predict behaviour in the wild? A study with pharmaceutical pollutants
M.G. Bertram, Monash University / Biological Sciences; M. Saaristio, J.M. Martin, T.E. Ecker, C.P. Johnstone, B.B. Wong, Monash University / School of Biological Sciences; A. Sundelin, J. Fick, Umea University / Department of Chemistry; A. Alarntä, Swedish University of Agricultural Sciences SLU / Department of Wildlife, Fish & Environmental Studies
Concern over the impact that pharmaceuticals have on wild aquatic organisms has increased over the past decade. Laboratory studies have shown that pharmaceuticals can cause sub-lethal changes to animal behaviour and physiology; however, few studies have addressed whether effects documented in the laboratory extend to the natural environment. We exposed fish to one of two pharmaceuticals (temazepam and ibersartan) commonly detected in the environment at two doses (80 ng/L, 1500 ng/L) - 200%, for dairy products 40% for wood products 50-300%, and for primary plastics 2-15%. The model is location agnostic and can be applied to any decision-making context concerning long-term effects of small-scale changes. The iLUC framework is now integrated in the global EXIOBASE model, thus accounting for all crops in all countries in the world. It has already been applied to more than 50 LCA studies and on several product categories.
The capacity of pharmaceutical pollution to alter behaviour in wildlife is of increasing concern to the scientific community. A major pathway of these contaminants into the environment is the treatment of livestock with hormonal growth promotants (HGPS), highly potent veterinary pharmaceuticals that can enter aquatic ecosystems via effluent runoff. Hormonal growth promotants are designed to have biological effects at low doses, often act on physiological pathways that are evolutionarily conserved across taxa, and have repeatedly been detected in ecosystems worldwide. However, despite being shown to cause altered development, reproduction and morphology in various non-target species, relatively little is known about the potential of HGPS to alter ecologically important behaviours, especially across multiple contexts. Here, we investigated the effects of short-term (24 h) and field-detected levels (4 h) of trenbolone—a potent growth-promoting veterinary pharmaceutical repeatedly detected in freshwater systems—on a suite of ecologically important behaviours in female eastern mosquitofish (Gambusia holbrooki). We found that fish exposed to 17β-trenbolone were more active and exploratory in a novel environment (i.e. maze arena), while boldness was not significantly affected. Further, when tested for sociability, exposed fish were again more active and exploratory, and spent less time associating with a shool of stimulus (i.e. unexposed) conspecific females. Lastly, when assayed for foraging behaviour, exposed fish spent a greater total amount of time within a foraging zone containing an array of prey items (chironomid larvae) than did unexposed fish, entered this zone more frequently, and were more likely to feed. Further, a grazing rate of chironomids on three microalgae species, independently. Therefore, although treatment-induced increases in foraging behaviour were dependent on female size. Taken together, these findings highlight the potential for sub-lethal levels of veterinary pharmaceuticals detected in the environment to alter sensitive behavioural processes in wildlife across multiple contexts, with possible ecological and evolutionary implications for exposed populations.

202 Selective grazing behaviour of chironomids between three microalgal species under pesticide pressure
J.N. Henry-Ornandi, Irstea / EABX-CARMA; C.N. Doose, INRS - Centre Eau Terre Environnement; B. CHAUMET, Irstea; N. Mazeli, Irstea Bordeaux / UR EABX; N. Majdi, Ecolab / UMR 5245 CNRS; J. Vedrenne, S. Morin, Irstea Bordeaux / UR EABX; W. Traunspurger, Bielefeld University / Animal Ecology. The herbicide diuron and the insecticide imidacloprid are among the most frequently detected pesticides in French rivers, and each is known to affect many aquatic organisms. However, it is less examined whether and how both pesticides together might affect chironomids. For this study, we used the grazing rate of chironomids on three microalgal species, independently. Therefore, two diatom species, Gomphomena gracile (two different morphotype: normal (GG) and teratogen (GT)) and Planolithium lanceolatum (PL), and one green algae Pseudokirchneriella subcapitata (PS) were offered as food, during 24h. Protein and lipid contents in microalgae were analysed subsequently. Each pesticide condition elicited a different grazing rate in chironomids with regards to algal species and their nutritional quality, with a general preference for Gomphomena gracile with teratogen shape and Pseudokirchneriella subcapitata. In a second experiment (cafeteria), food selectivity of chironomids was determined under similar contamination conditions during 4h: Under diuron, larvae switched equally among microalgae, then were as mobile as in the control without pesticide. However, imidacloprid and the pesticide mixture condition altered chironomids’ movements and grazing behaviour. In this experiment, we highlighted that chironomids feeding behaviour and food preferences are impacted by pesticides. Herbicide and insecticide exposure, alone or in combination, had contrasting effects on grazing, both directly on the larvae or indirectly (food selectivity according to its quality). Our study illustrates the value of considering the impacts of toxicants on target and non-target organisms across trophic levels to improve ecotoxicological risk assessment in an ecosystemic perspective.

203 Environmental levels of anxiolytic pharmaceuticals after migration of Atlantic salmon in both lab and field
T. van der Klis, Umea University / Department of Ecology and Environmental Science; G. Hellström, Swedish University of Agricultural Sciences SLU; J. Klämlander, Umea University / Ecology and Environmental Science; F. Finn, Umea University; A. Lagesson, Umea University / Department of Ecology and Environmental Science; M. Jonson, Umea University; J. Fick, Umea University / Department of Chemistry. Humans consume more pharmaceuticals than ever and consumption is set to rise. As a consequence, increasing amounts of pharmaceuticals are released into waterways worldwide with virtually no knowledge of how they might affect aquatic ecosystems. Some conspicuous effects of these emerging contaminants are already evident including the feminization of fish by contraceptive residue. However, recent work suggests that important effects of pharmaceuticals in aquatic environments are much more widespread than currently believed, and that these effects may result in major changes in species interactions, population survival and ecosystem functioning. In several earlier laboratory studies, we have shown that concentrations of pharmaceuticals presently found in waterways alter important behavioural traits in both aquatic macroinvertebrates and fish, and that this in turn affects both feeding efficiency and predation risk. These results suggest that pharmaceutical contamination of aquatic environments may change species interactions, in particular predator-prey interactions, with severe ecosystem-effects as potential consequence. Recently our research focus has turned towards realistic large-scale studies in lakes and rivers using acoustic telemetry to test if findings from the lab also hold under field conditions. Here I present results from one such study comparing effects of environmental levels of the anxiolytic pharmaceutical Oxazepam on migration pattern of Atlantic salmon (Salmo salar) in the lab and the field. In the lab, salmon exposed to the drug migrated approximately twice as fast as unexposed salmon and the subsequent field-study generated similar results, validating the results found in the lab. This pharmaceutically induced change in migration-intensity has the potential to be a key determinant between survival and mortality of salmon individuals and as such important for population persistence as migration intensity is believed to be adapted to the environmental conditions of the river in question. The overall finding of recent studies suggests that effects of pharmaceutical contamination of natural systems might be much more widespread than we predict based on conventional ecotoxicological tests.

204 Can personality influence the response of fish to environmental contaminants?
M. Oliveira, University of Aveiro; M. Sampaio, T. Santos, University of Aveiro / Biology Department & CESAM; A.L. Machado, University of Aveiro / CESAM Department of Biology; M. Barbosa, Department of Biology & CESAM - University of Aveiro / Biology; I. Domingues, University of Aveiro / CESAM Department of Biology. Stress is a determinant factor reducing animal welfare. Currently, it is recognized that animals react differently as a function of their personalities, or stress coping styles (i.e. consistency in behavioural and physiological responses across time or contexts). However, the role of personality in modulating individual response to environmental contaminants has received limited attention, despite the recognition that personality traits associated with a shy-bold continuum play an important role in animal fitness. Knowing that pharmaceuticals can interfere with personality, one question arises: what is the role of personality on animals’ response to stressors? In this research, we aimed to determine whether personality, assessed by object neophobia, exploration in a novel environment, and boldness, is more active on average but fluoxetine treated birds showed no increase in activity, or indirectly (food selectivity according to its quality). We found that fish exposed to 17β-trenbolone were more active and exploratory in a novel environment (i.e. maze arena), while boldness was not significantly affected. Further, when tested for sociability, exposed fish were again more active and exploratory, and spent less time associating with a shool of stimulus (i.e. unexposed) conspecific females. Lastly, when assayed for foraging behaviour, exposed fish spent a greater total amount of time within a foraging zone containing an array of prey items (chironomid larvae) than did unexposed fish, entered this zone more frequently, and were more likely to feed. Further, a grazing rate of chironomids on three microalgal species, independently. Therefore, although treatment-induced increases in foraging behaviour were dependent on female size. Taken together, these findings highlight the potential for sub-lethal levels of veterinary pharmaceuticals detected in the environment to alter sensitive behavioural processes in wildlife across multiple contexts, with possible ecological and evolutionary implications for exposed populations.

Several species of bird are known to forage directly on invertebrates at wastewater treatment plants (WWTPs), representing an exposure route to a range of contaminants including pharmaceuticals. The selective serotonin reuptake inhibitor (SSRI) fluoxetine is heavily used and could induce indirect effects on WWTPs. Since fluoxetine is commonly prescribed for anxiety, we hypothesised that the antidepressant could modulate anxiety behaviour and physiology in exposed birds. Anxiety is an important state which arises in response to a real or perceived threat, enabling the individual to respond appropriately. Contaminants with the potential to alter anxiety-related behaviours are thus of concern to wildlife. We conducted a study to investigate the potential for sex-dependent differences in exposure to a relevant concentration of fluoxetine in a model songbird, the Eurasian starling (Sturnus vulgaris). We used a combination of behavioural and physiological endpoints to assess the effects of fluoxetine, specifically: 1) object neophobia, 2) exploration in a novel environment, 3) activity levels, 4) faecal corticosterone (CORT) metabolite concentration; and 5) leg skin temperature. Compared with pre-treatment data, fluoxetine-treated birds became less neophobic on average after six weeks of dosing, indicating a decrease in anxiety behaviour. There was no such reduction in neophobia in the control group. After six weeks of dosing, control birds became more active on average but fluoxetine-treated birds showed no increase in activity,
indicating increased lethargy in the fluoxetine birds relative to controls. There was no clear effect of treatment on exploratory tendency. Finally, infrared thermography showed that fluoxetine-treated birds had significantly colder legs compared with controls. This indicated that, as observed in humans, fluoxetine causes vasodilatation, which in birds will affect the ability to thermoregulate. This study provides further evidence that low, environmentally relevant concentrations of pharmaceuticals can cause subtle changes to behaviour and physiology that are predicted to impair the capacity of wildlife to respond appropriately to environmental changes.

Can trends in wildlife populations revolutionise our understanding of the impacts of chemicals on the environment?

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Does single compound risk assessment protect from mixture effects and multiple stress?

P. Von der Ohe, UBA - Federal Environment Agency / IV 2.2 Pharmaceuticals

There is clear evidence that stress from anthropogenic activities can have profound local and regional effects on aquatic communities. To what extent chemicals are responsible remains largely unknown. The question whether a single compound risk assessment can protect from further deterioration of our water resources is discussed in the light of current mixture toxicity frameworks and multiple stress considerations. Here we present a European wide risk assessment of organic chemicals, based on regulatory monitoring data at about 6,000 monitoring sites available from the Dutch Environment Agency (EEA). For the more than 600 mostly industrial substances, including many detergent ingredients such as benzotriazol, the available quality standards were collated or predicted from reliable QSAR models. Results showed that organic chemicals are likely to exert long-term effects on sensitive species in more than 9% of the sampling sites with multi-year samplings. In this study, we analyzed the potential cumulative effects of multiple exceedances of the PNEC (predicted no-effect concentration) EC50 beyond which species abundance starts changing when toxic chemical stress, but differing habitat quality and b) similar habitat quality but differing chemical stress. The results indicate that in direct comparison, chemical stress induced larger effects as compared to habitat degradation. Our results therefore clearly indicate that chemical pollution is still a large-scale environmental problem that requires far-reaching, holistic mitigation measures to preserve and restore ecosystem health. It also suggests that current risk assessment approaches on a substance by substance bases are likely not protective for the environment.

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Threshold trends in wildlife taxa: challenging and evaluating our chemical- and environmental risk assessments of chemicals and their mixtures

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“Big data” are a potential goldmine for studying and contextualising chemical stress, but differing habitat quality and b) similar habitat quality but differing chemical stress. The results indicate that in direct comparison, chemical stress induced larger effects as compared to habitat degradation. Our results therefore clearly indicate that chemical pollution is still a large-scale environmental problem that requires far-reaching, holistic mitigation measures to preserve and restore ecosystem health. It also suggests that current risk assessment approaches on a substance by substance bases are likely not protective for the environment.

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How much do improvements in wastewater treatment benefit downstream macroinvertebrate populations?

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The River Ray, which is a small tributary of the Thames (UK), offers an unrivalled opportunity to examine the impact of changes in wastewater treatment on the resident aquatic wildlife. This opportunity exists because the waste from the 170,000 plus people of Swindon discharges into this small 12 km long waterbody, such that the downstream mean annual flow is composed of 65-80% treated effluent. The downstream monitoring sites showed a sustained improvement in macroinvertebrate diversity starting from 1991 onwards. This sustained improvement for macroinvertebrates coincided with a 10-fold drop in ammonia, halving of biodegradable organics, (BOD) and improvement in dissolved oxygen associated with the conversion of the Swindon plant from trickling filter to nitrifying activated sludge. There were no dramatic changes in metal concentrations over the key early 1990s’ period unlike the main sanitary determinants. Whilst there was no change in overall flows, winter water temperatures downstream of Swindon rose over the course of the 30 year monitoring period. We could not identify how this benefited the macroinvertebrate assemblage from the application of tertiary granular activated charcoal treatment (GAC) lasting from 2008 to 2014. No new macroinvertebrate families appeared during this period. The steady improvement in macroinvertebrate diversity in an effluent dominated river implies that current chemicals in domestic wastewater are not noticeably harmful to these organisms. This implied that provided we can achieve a 90% percentile BOD below 5 mg/L, NH below 0.6 mg/L and DO above 60% saturation, the downstream macroinvertebrate diversity will steadily improve (within the limits of habitat suitability). The small river has a population of stickleback and other fish but their changes in abundance are not known. The recovery of macroinvertebrate diversity is slow and has not yet returned to the expected range for such a river, however, it was noted that the habitat is not ideal. This project has demonstrated the immense power and value of consistent long-term parallel chemical and wildlife monitoring. Andrew C. Johnson, François Edwards, Monika D. Jürgens, Helen Vincent Centre for Ecology and Hydrology, Wallingford, Oxfordshire, OX10 0E8, UK ajoo@ceh.ac.uk Keywords: macroinvertebrates, biodiversity, wastewater, GAC Track 7, Session 7.2 Preference Platform

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Biometric parameters of the bream (Abramis brama) as indicators for long-term changes in environmental quality - results from the German ESB

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Fish health depends upon the macroinvertebrate community from the application of tertiary granular activated charcoal treatment. Changes in physical condition of fish may therefore be attributed to changes in environmental quality. Based on time series of 20 years of biometric data of bream from multiple sampling sites of the German environmental specimen bank (ESB), we assessed which biometric parameters and indices of bream are suitable indicators for long-term changes in fish health and environmental quality. The length and weight of individuals of a defined age, hepatosomatic index and wet restriction the condition factor and lipid content of bream are reliable indicators for long-term changes of fish health and hence hint at long-term changes of environmental quality. We show examples for current trends of these indicators in German river systems. Our results confirm the high value of biometric parameters for monitoring of long-term changes in state and quality of aquatic ecosystems.

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The burden of being a slow-life cycle species: freshwater fish population dynamics in France, correlations to species life traits and implications in ecotoxicology

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Population dynamics of aquatic species and ultimately their population growth rate (λ), must be known to properly define species conservation status and plan appropriate conservation strategies. It also offers a tool for ecosystem research and conservation. Particular interest is in the potential for inter-specific variations in demographic and life history traits influence population dynamics. For this purpose, we implemented an integrative approach focused on 18 common freshwater fish species representing 94% of fish abundance and 88% of fish biomass sampled since 1990 in 546 monitored sites in France. Abundance and biomass growth rates were estimated with space-models and fish length trends with quantile regressions. To further study correlations between fish abundance, biomass, fish-length trends and fish life traits (life history strategies, species trophic position, habitat preferences and thermal tolerance) we performed multivariate analyses. The present work demonstrates that during the last decades, 10 species
exhibited significant decline in abundance, 2 species were in expansion and fish abundance remained stable for 6 species. The correlation between biomass and abundance growth rates was also very high (R²=0.93). The intra-specific trends in fish length over the studied period also showed a severe decrease among the largest individuals (quantile 0.75 and 0.90) and was correlated to severe biomass decline in several species. This result reflect progressive alterations in the population size / age structure suggesting that a decrease in growth and survival might be responsible of the slow declines observed. Among the demographical and ecological traits we investigated, generation time and fish maximum length were the most correlated to species population growth rates indicating the decline of slow generation time species. These results are discussed with regards to global pressures which could explain large scale decline of periodic species with a focus on chemical pollution which could explain body length decrease, juvenile and adult survival alterations due to micropollutant exposures. Further attention will be paid to discuss conservation measures and life stages which should be protected in priority to favor periodic species recovery.

211 The use of natural historical collections to reconstruct temporal trends of the exposure to major contaminants in different white-tailed eagle (Haliaeetus albicilla) populations

J. Sun, Antwerp university / Department of Biology; J.O. Bustnes, Norwegian Institute for Nature Research / Fram Centre; A. Covaci, University of Antwerp, Toxicological Center / Toxicological Centre Dep of Pharmaceutical Sciences; B. Hipsø, Swedish Environmental Monitoring; G. Malavannan, University of Antwerp / Toxicological Center; J. Søndergaard, Aarhus University / Arctic Research Centre; K. Thorup, A. Tøttrup, Natural History Museum of Denmark; M. Eens, University of Antwerp / Department of Biology; I. Eulaers, Aarhus University / Department of Bioscience Understanding temporal trends of persistent contaminants in wildlife is critical for the assessment of the pattern here observed, present and future health risks. We have established a retrospective examination of Mercury (Hg) and several organohalogen contaminants (OHCs) in Swedish, Norwegian and Greenlandic white-tailed eagle (Haliaeetus albicilla) using body feathers obtained from natural history and ornithologist collections. We analyzed feathers for Hg, polychlorinated biphenyls (PCBs) and polybrominated diphenyl ethers (PBDEs), as well stable carbon and nitrogen isotope ratios. The aims of the present study were: 1) to retrospectively reconstruct temporal trends of major contaminants in relation to anthropogenic emissions and regulations, 2) to interpret how dietary habits (using stable isotope proxies) may have affected the temporal trends, and 3) to document a potential spatial gradient from high (Sweden) to low (Greenland) anthropogenic contaminant input. Strong evidence for spatiotemporal patterns was found based on preliminary results for Hg and OHCs in the Swedish and Norwegian populations. Swedish white-tailed eagle feathers showed concentrations associated with adverse physiological effects, thus supporting the earlier observed dramatic decline in productivity. Hg concentrations were much lower in the Norwegian feathers, while the peak PCB exposure is of the same order of magnitude as the average PCB exposure in the Swedish and Norwegian time series after 1970s. We did not detect PBDEs before 1970 in the Swedish or Norwegian time series, although concentrations increased steeply during the 1980s, decreasing trends were observed afterwards. Hg concentrations in the Greenlandic time series were stable before 1940, but showed an increasing trend from 1980 to 2013. These results suggest that changes in historical and recent anthropogenic emissions strongly shape temporal changes of persistent contaminants in avian top predators. In addition, the relative interplay between local versus long range transport have likely affected the spatial pattern of contamination levels. The here presented preliminary results will be complemented with data for the Greenlandic populations as well as emission and stable isotope data to disentangle the relative effects of dietary habits and anthropogenic contaminant sources.

Environmental effects of metals: Improvements to risk assessment by considering speciation and bioavailability (I)

212 Findings of a SETAC Technical Workshop on Bioavailability-Based Water Quality Criteria

C.E. Schlekat, NIPERA; K. Gallagher, U.S. EPA / Office of Water; B.A. Stubblefield, Oregon State University / Environmental and Molecular Toxicology A workshop entitled “Technical Workshop on Bioavailability-based Water Quality Criteria” was held under the auspices of the Society of Environmental Toxicology and Chemistry on 3-8 December 2017 in Pensacola, Florida, USA. The goal of the workshop was to evaluate the state-of-the-science regarding metals bioavailability models and their use from a regulatory perspective. A second, but equally important, goal was to provide a scientific forum for discussions that could inform the further incorporation of bioavailability concepts into regulatory approaches for determining heavy metal risk. The workshop focused on different aspects of the state-of-the-science regarding the ability to accurately predict the toxicity of metals in the environment to aquatic organisms has increased rapidly in recent years. Workshop participants reviewed the current state-of-the-science on aquatic bioavailability information for metals and on methods to model bioavailability under a range of environmental conditions. Participants also discussed the technical challenges associated with applying bioavailability-based approaches, especially in a regulatory context. The outcome of the workshop will support expanded incorporated of metal bioavailability information into global regulatory frameworks. Approximately 40 invited scientists representing industry, governments, and academia participated in the workshop. Participants were divided into five working groups, each addressing one of the following meeting objectives: Review of the state-of-the-science regarding the issue of metal bioavailability and toxicity to aquatic organisms, Determine the extent to which available biotic ligand models (BLM/multi-linear regression (MLR)-based models/other alternative approaches) offer a means to model bioavailability under a range of conditions and to which they are protective of aquatic life. Develop technical information regarding the type and quantity of data necessary to develop and apply bioavailability-based aquatic toxicity models, Develop recommendations for approaches to validate bioavailability-based models, and Provide recommendations for suggested measures of acceptability for bioavailability-based aquatic effects models. Workshop findings will be presented and will later be published in the form of a SETAC “Summary document” and a series of manuscripts to be submitted to a SETAC journal.

213 Modifying factors for nickel speciation and toxicity in seawater

W. Chen, Wilfrid Laurier University; S. Sherman, Wilfrid Laurier University / Biology; J. McGeer, Wilfrid Laurier University / Department of Biology; R.C. Santore, Windward Environmental, LLC; T. Blewett, University of Alberta; G. Merrington, WCA Environment Limited; D. Smith, Wilfrid Laurier University / Department of Chemistry Nickel (Ni) toxicity in seawater is of increasing concern because of coastal Ni mining and processing activities. Determining Ni speciation is vital to understanding and predicting Ni toxicity and ultimately for bioavailability based nickel risk assessment. Application of existing freshwater bioavailability based approaches for nickel in salt water predicts negligible binding of Ni to dissolved organic matter, but there are several examples of toxicity tests in natural seawater that are protective compared to artificial seawater control samples. Determining Ni speciation is vital to understanding and predicting Ni toxicity and ultimately for bioavailability based nickel risk assessment. Application of existing freshwater bioavailability based approaches for nickel in salt water predicts negligible binding of Ni to dissolved organic matter, but there are several examples of toxicity tests in natural seawater that are protective compared to artificial seawater control samples. Determining Ni speciation is vital to understanding and predicting Ni toxicity and ultimately for bioavailability based nickel risk assessment. Application of existing freshwater bioavailability based approaches for nickel in salt water predicts negligible binding of Ni to dissolved organic matter, but there are several examples of toxicity tests in natural seawater that are protective compared to artificial seawater control samples. Determining Ni speciation is vital to understanding and predicting Ni toxicity and ultimately for bioavailability based nickel risk assessment. Application of existing freshwater bioavailability based approaches for nickel in salt water predicts negligible binding of Ni to dissolved organic matter, but there are several examples of toxicity tests in natural seawater that are protective compared to artificial seawater control samples. Determining Ni speciation is vital to understanding and predicting Ni toxicity and ultimately for bioavailability based nickel risk assessment.

Acute bioavailability models for nickel: Development and regulatory application

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Chronic bioavailability models for nickel are well-established. For example, the Annual Average Ni Environmental Quality Standard (AA-EQS) under the European Union’s (EU) Water Framework Directive (WFD) is based on normalization of chronic Ni ecotoxicity data using chronic Ni bioavailability models. Acute Ni models have been developed for invertebrates, fish, and algae, but data to develop acute Ni ecotoxicity bioavailability models are limited. However, given that acute Ni toxicity can vary by greater than 17-fold for aquatic invertebrate species tested in variable water chemistries, normalization of acute Ni ecotoxicity data is important for determination of Maximum Allowable Concentrations (MAC) under the WFD. The goal of this study was to test if the existing acute Ni bioavailability models can be used to predict acute Ni toxicity to both model and non-model species and to demonstrate how they can be applied to derived MAC values. Acute Ni ecotoxicity data (eight species from 13 different studies) were identified from the literature. Data were accepted for analysis if acute Ni toxicity for a species was tested in >2 test waters differing in physico-chemistry.
Acute Ni bioavailability models (3 invertebrate models, 2 algae models, and 1 fish model) were used to evaluate the ecotoxicity data. To simplify the normalization process, an “average animal” bioavailability model was developed using a weighted average of parameters for existing models. Because crustaceans are typically among the most sensitive organisms to Ni exposure, and because the fish model did not capture Ni effects on acute Ni toxicity to crustaceans very well, an “average crustacean” model was developed. The model parameters for 3 crustacean model weights were used to construct the “average crustacean” model. Both the “average animal” and “average crustacean” models reduced intraspecies variability considerably among the available Ni ecotoxicity data. For example, the “average animal” model predicted 98% of the 193 individual acute ecotoxicity data points within a 3-fold error, and 90% within a 2-fold error. The “average crustacean” model performed similarly. Although this model clearly showed a better ability to predict the effect of pH on Ni toxicity to cladocerans. The models were applied to an acute Ni ecotoxicity dataset to derive bioavailability-based MAC for European water bodies with typical ranges of water chemistry.

215 Bioavailability and bioaccumulation of uranium: From lab experiment to modelling

A. Husson, Mines ParisTech / Hydrodynamics and reactions team (HR); M. Learmers, Vrije universiteit Brussel / Department Analytical, Environmental and Geochemistry; M. Descostes, AREVA Mines / R&D; V. Lagneau, Mines ParisTech PSL, Research University (FR (Geosciences and health of rocks (HR); V. Lagneau, Mines ParisTech / Hydrodynamics and reactions team (HR) Environmental regulations are becoming more and more ecosystems-oriented. In this respect, the habitat’s faunal biodiversity is targeted, meaning the contaminant’s bioavailability is taken into account. The latter depends on the geographical conditions of the aquatic system, which define the chemical speciation. For several elements, Environmental Quality Standards (EQS) and Sediment Quality Standards (SQR); have been proposed. For non-priority elements such as Uranium, EQS and SQS should be supplemented with national standards. As the bioavailable fraction in sediments is dependent on geochemical factors and sediment mineralogy, it is important to investigate the role of sediment composition on the bioaccumulation of uranium in benthic organisms. In the present study, laboratory experiments were performed on the bioaccumulation of uranium in the larvae of the non-biting midge Chironomus riparius exposed to specific mineralogical phases (pure Quartz (Q); 10% Kaolin/90% Q, 10% Silticite/90% Q, 10% Ferrihydrite (FOH)/90% Q and a mixture of the 4 mineral phases (3.3% Kaolin/3.3% Silticite/3.3% (FOH) and 90% Q) spiked with uranium at two different concentration levels. During a ten days’ exposure experiment, the uptake of uranium in the chironomid was investigated and the concentrations of uranium in sediment, overlying water, pore water were measured as well as the composition of major ions and physicochemical parameters. Diffusive Gradients in Thin Films (DGT) devices were deployed simultaneously to investigate the relationship with the uptake of uranium in the chironomid larvae Sediment to porewater partition coefficients (Kp) range from 10 L/Kg for quartz to 60000 L/Kg for FOH. After ten days exposure, the highest Biota Sediment Accumulation Factor (BSAF) was spotted for pure Quartz (406) and Ferrihydrite (9) and Silticite (8) is the lowest for the mixed composite sediment (1). DGT labs show that the porewater concentrations account for 70–100% of the uranium in porewater for all mineral phases except the quartz, where Cext only accounts for 10% of the uranium in porewater. Uranium accumulation on the DGT units is correlated with the accumulation in the chironomids. Results obtained by the highest BSAF experiment are used in code (CUMINS) to model the uranium sorption behaviour and chemical speciation in the aqueous phase. These results are compared with the proposed regulations by IRSN on uranium bioavailable chemical species.

216 Empirical Investigations into the Toxicity and Bioavailability of Aluminium to Aquatic Species

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217 Main factors responsible for the environmental degradation of rivers in a basin dedicated to gold mining using ecological predictive models. Case study Ponce Enríquez.


The irreversible effects that the environment has suffered due to anthropogenic activity has reduced the availability of water for living beings, both in terms of quantity and quality. Industrial, agricultural, mining and urban development activities have in many cases led to the generation of pollutant discharges that threaten the health of our ecosystems. The impacts of mining activity on aquatic ecosystems have been widely documented, reporting the deterioration of water quality, the impact of biodiversity, as well as the release of heavy metals of potential accumulation in organisms and subsequent biomagnification through the food chain. Although it is known that non-technical mining activity affects the environment, it is necessary to identify and prioritize those factors related to mining activity that have a greater impact on the ecosystem (e.g. extraction, cyanidation, amalgamation). The identification of these factors would allow environmental control authorities to prioritize management actions focused on those parameters with the greatest impact, thus mitigating the impact of this activity on aquatic ecosystems. To illustrate this, the present study conducted in the Ponce Enríquez area seeks, through the construction of predictive models based on decision trees, to discriminate the main factors responsible for the environmental degradation observed in rivers and streams in the study area. In order to determine the environmental quality of the sites visited, the application of the BMWP / Col index was used, which is based on the structure of the macroinvertebrate community present. Additionally, a set of environmental variables of water and sediment quality were used to develop and validate the predictive model. The model was exposed to specific mineralogical phases (pure Quartz (Q), 10% Kaolin/90% Q, 10% Silticite/90% Q, 10% Ferrihydrite (FOH)/90% Q and a mixture of the 4 mineral phases (3.3% Kaolin/3.3% Silticite/3.3% (FOH) and 90% Q) spiked with uranium at two different concentration levels. During a ten days’ exposure experiment, the uptake of uranium in the chironomid was investigated and the concentrations of uranium in sediment, overlying water, pore water were measured as well as the composition of major ions and physicochemical parameters. Diffusive Gradients in Thin Films (DGT) devices were deployed simultaneously to investigate the relationship with the uptake of uranium in the chironomid larvae Sediment to porewater partition coefficients (Kp) range from 10 L/Kg for quartz to 60000 L/Kg for FOH. After ten days exposure, the highest Biota Sediment Accumulation Factor (BSAF) was spotted for pure Quartz (406) and Ferrihydrite (9) and Silticite (8) is the lowest for the mixed composite sediment (1). DGT labs show that the porewater concentrations account for 70–100% of the uranium in porewater for all mineral phases except the quartz, where Cext only accounts for 10% of the uranium in porewater. Uranium accumulation on the DGT units is correlated with the accumulation in the chironomids. Results obtained by the highest BSAF experiment are used in code (CUMINS) to model the uranium sorption behaviour and chemical speciation in the aqueous phase. These results are compared with the proposed regulations by IRSN on uranium bioavailable chemical species.

218 Closing the gap between small and smaller: Towards a framework to analyse nano- and microplastics in aquatic ecosystems

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Detecting nanoplastics and determining actual concentrations and sizes of plastic particles present in the environment is essential to assess the risks that are posed by plastic particles. Microplastics have been detected globally in various aquatic ecosystems. The determination of nanoplastics is hampered due to the high methodological challenges. But yet, its formation was proven experimentally and, for the first time, its presence in marine surface water samples confirmed. Building on those results, we further propose a framework that is able to consistently determine a broad size spectrum of plastic particles in an aquatic environmental sample. The results from initial tests confirm the general applicability of individual techniques to, firstly, sample and, secondly, detect plastic sizes and polymer types. To obtain representative results, a sampling strategy is needed to concentrate plastic particles. Crossflow ultrafiltration is applied to concentrate nanoplastics from 100 into 0.5 L and yields in a reproducible particle recovery of 54.2 ± 2%. Microplastics are detected using FTIR-microscopy which is limited to a minimum particle size of 28 µm. For nanoplastics field- flow fractionation, that reveals information on the particle sizes, and pyrolysis GC-MS, that is used to identify the polymer types, are
applied. Under the given settings the latter requires a mass of approximately 100 ng to identify polystyrene in an environmental sample by which this technique seems promising for the detection of nanoplastics. The pre-concentration by crossflow ultrafiltration reduces the determined detection limits, and enables the identification of polystyrene for an original concentration of 20 μg L⁻¹ in an aqueous samples. Finally, we propose an approach to estimate polymer masses based on the two-dimensional microparticle fingerprinting technique, evaluating the analysis with FTIR-microscopy. By this, the results of spectroscopic and thermal degradation analyses (e.g. pyrolysis GC-MS) can be combined and compared.

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Trace particulate plastic analysis in environmental systems: synthesis and utilisation of chemical tracers and microplastic and microfibres: A. Lusher, F. Schmidt, M. Schmiededer, Eawag – Swiss Federal Institute of Aquatic Science and Technology; D.M. Mitran, Eawag - Swiss Federal Institute of Aquatic Science and Technology / Environmental Risk and Management Research on particulate plastic (nano- and microplastic particles and fibers) and their distribution in the environment has intensified in recent years; but truly quantitative analysis, even at the bench scale, has remained elusive in part due to the analytical difficulties in detection. Synthesizing plastic particles with a metallic, chemically entrapped tracer can provide a robust way to more easily, accurately and quantitatively detect particulate plastic in complex environmental and biological media. In this study, a suite of methods to synthesize a variety of particulate plastics of various sizes (100 nm to 1 mm), surface morphologies/charges and polymers (polystyrene, polyester, polypropylene and low density polyethylene). Each variant has an embedded metallic fingerprint (Pd, In, Au; approximately 0.5% metal/wt) which can be used to detect plastic by common analytical techniques, such as ICP-MS. To highlight the utility of this approach, dilute concentrations of particulate plastic were measured in various media including river water, municipal wastewater treatment plant effluent and wastewater sludge. Nanoplastics, for example, were detected three orders of magnitude lower than the equivalent concentration than similar particles with a fluorescence label. After ensuring the metal tracer was stable over time, digestion methods were developed which took into consideration the specific polymer and metal in question to achieve a minimum of 85% recovery in every matrix. Furthermore, the particulate plastics synthesized were spiked into nixed aqueous batch experiments, representing different stages of a municipal wastewater treatment plant to determine how various particulate plastics move through the system. In this way, some conclusions could be drawn concerning particle behaviour, aggregation and the likelihood of microplastics and the potential for microplastics to be discharged to freshwater within wastewater treatment effluent. Beyond the case study specifically highlighted here, these metal laden particulate plastics are a promising tool for studying fate and transport, and interactions with organisms at trace concentrations. By using these materials, bench scale and pilot scale studies can be used as a bridge to understand the environmental processes that dominate (particulate) plastic fate, transport and interactions with biota until analytical techniques to measure native particulate plastics of small sizes are developed. The TED approach is currently a step analytical procedure which consists of a thermal desorption (TD)-gas chromatography mass spectrometry (GC-MS). The TED sample preparation and the analytical tool, the so-called TED MS, is a two-step analytical procedure which consists of a thermal extraction where the sample is annealed and characteristic degradation products of the polymers are collected on a solid phase. Afterwards these products are analysed using GC-MS [5]. The developed fractional filtration for sampling and the TED approach for detection were used for quantitative analysis to screen the waste water influent and effluent of a Berlin waste water treatment plant for the most relevant polymers, polystyrene (PE), polypropylene (PP), polyethylene (PS), polyethylene terephthalate (PET) and polyamide (PA). The results of the study revealed that the polymers PE, PP and PS were detected in the effluent, and PE and PS were found in the raw waste water of the sewage treatment plant in Ruhleben, Berlin. Differences in polymer types and amounts were detected at different sampling dates and within different sieve fractions. Much higher amounts of polymers were observed in the raw waste water. The peak areas of the decomposition products, used for quantification of the polymers, were adjusted using so-called response factors since the TED-GC-MS method is more sensitive for PP and PS than for PE. It has been shown that PE is the most dominant polymer in the samples. Comparing the masses of polymers in the effluent and in the raw sewage, a removal of 97 % of the polymers in the water treatment plant can be assumed. These results are consistent with the literature where removal rates between 98-99 % were described [6-7].

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Soil and sludge: A time and cost-effective method for extracting microplastics from complex, organic-rich environmental matrices

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There is very little existing information on microplastics in organic-rich substrates such as soil or sewage sludge. The organic components, as well as the complexity of the solid matrix, complicates the extraction process. No standardised methodology has thus far emerged. This study aims to establish an effective extraction technique appropriate for the monitoring of microplastic contamination in soil and sludge samples. Four main protocols (including two temperature and concentration variants) were tested for the removal of organic material followed by a density separation process. This approach was selected to afford comparability with existing sediment microplastic analyses. The selected reagents were: peroxide oxidation (60°C, 70°C), Fenton’s reagent, NaOH (1 M, 10 M) and KOH. The methods testing procedure was split into three phases: 1. Effect of reagents on test polymers; 2. Efficacy of reagents in reducing organic matter content in soil and sludge; and 3. Extractions surveying the incorporation of sludge samples including density separation. 7 reference polymers were used: HDPE, LDPE, PP, PS, PET, PC and PA-66, which account for >70% of global plastic demand. Degradation was observed for several of the reagents and selected polymers. NaOH treatments caused the most significant changes, including degradation of PET and PC. Small degradative changes occurred as a result of peroxide oxidation and KOH treatments. Fenton’s reagent, NaOH and KOH treatments were not effective in sufficiently reducing organic material whilst peroxide oxidation and Fenton’s reagent removed the majority of organic matter. Based on these results, Fenton’s reagent was identified as the optimum treatment and was further optimised. Phase 3 tested revealed high extraction efficiencies for this protocol. This study showcases a time- and cost-effective approach suitable for application in routine monitoring of complex environmental substrates whilst facilitating the collection of important particle information.

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Mapping microplastics in sludge during a country-wide investigation of wastewater treatment plants

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Incorporation of anthropogenic particles into sludge has been highlighted as a major route for the transport of pollutants into the environment. Here we present the results of a countrywide survey investigating the incorporation of microplastics into sludge from domestic wastewater treatment plants which operate different waste water and sludge treatment technologies. The main objective of this project was to characterize, map and compare results between different types of wastewater treatment plants. Samples were extracted using organic matter removal followed by a density separation procedure following. Plastics were identified in sludge samples from all treatment plants investigated. The average, average microplastic concentration was 6 077 particles kg⁻¹ (d.w.) (1701 – 8 383) or 1 176 889 particles m⁻³ (470 270 – 3 394 274). Particles from sludge consisted of beads (37.6%), fragments (31.8%) fibres (28.9 %) and glitter (1.7 %). Most of the particles were clear in colour (41%). Ten percent of the overall particles extracted were tested using FT-IR. All particles (n= 60) were confirmed to be plastics. Polyethylene particles were the most common (30.5%) followed by polyethylene terephthalate (26.7%) and polypropylene (20.3%). 62% of plastics were extracted during the low density (1 g cm⁻³) separation steps and 38% were extracted at high density (1.8 g cm⁻³). Results between WWTPs were highly variable and there was no clear difference between average microplastic concentrations across the different treatment technologies. Based on the activities and results of this study on the application of sewage sludge in Norway, it can be estimated that approximately 446 billion microplastic particles are spread on agricultural soils, 27 billion microplastic particles are added to green areas and 112 billion microplastic particles are sent to soil producers per year. This equates to over 584 billion microplastics that are released into the environment via sewage sludge application each year in Norway.

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The Influence of Weathering on the Sinking Behaviour of Microplastic in Freshwater and all Surface Waters

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Key to understanding the fate of microplastic particles in fresh water are the changes they undergo as a result of various weathering processes, like biofilm growth, aggregation, UV exposure and physical stress. Some of these weathering
Air Pollution, Biomonitoring and Human Health (I)

224 Particulate matter in indoor academic environments: chemical composition, sources, infiltration from outdoor

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We present the results of the first part of an experimental study carried out in an academic building. Sampling was performed using Digitel® DA80 high volume samplers between November 2010 and April 2011 in three medium cities located in northern France, Dunkerque (Dk, coastal urban and industrial site), Boulogne-sur-mer (BL, coastal and urban site) and Saint-Omer (StO, inland urban and industrial site). PM2.5 composition was analyzed for major elements, trace elements and major species. Species concentrations were examined according to different ways including temporal evolution, concentration and pollution roses. The impact of such sources on major species and metal concentrations in PM2.5 was then quantified using a weighted non-negative matrix factorization based receptor model that considers constraints on chemical profiles (CW-NMF). NOx, SO2, NH3 and TC were found as the major components of PM2.5 (between 95% and 99%). Only significant differences were evidenced. Trace elements (Cr, Zn, Ni, As, Ag, Cd, Cu, Mn, Pb, V, Sn, Rh, Sr, Bi, Ba, Co, Sb and Tl) only correspond to 0.30% to 0.45% of the PM2.5 mass according to the sites. The CW-NMF model identified 8 common source profiles at the 3 sites: secondary nitrates, residential and road traffic, secondary sulfates, fresh and aged sea-salts, heavy fuel oil combustion, non-exhaust traffic and crustal. In addition, 4 industrial source profiles were identified in Dk and sulphuric emissions and sintering stack of an integrated steelworks, electric steelmaking and glassmaking activities. Despite their low contribution to PM2.5, such industrial sources were the main contributors of metals at the two sites.

225 Estimating the contribution of deposition in the total exposure to PAH's in order to derive save deposition reference values

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Partitioning of PAHs between the particulate and the gaseous phases strongly influences their fate and transport in the atmosphere and human exposure. Dry and wet deposition processes are major sources for PAHs in soil and crops. Our aim was to determine the proportion of the overall burden of environmental and dietary exposure to PAHs that is attributable to deposition in order to derive save deposition reference values. To this end, the fate and human exposure was modelled using the MERLIN-Expo, a software tool that allows to model lifetime exposure, integrating exposure through multiple pathways. Model simulations were based on recent yearly average concentrations in air and particulate matter (PM10) in Belgium. The conceptual model implemented, included inhalation, soil and dust ingestion as well as dietary exposure via the consumption of vegetables, meat and dairy products. Toddlers were chosen as the receptor as they are considered a vulnerable group. Dietary intake exposure to PAHs via crops was modelled using a plant uptake model representing leafy vegetables, fruits and grain, respectively. A cattle model taking its inputs from a grass and maize model was used to calculate concentrations in meat and dairy products. Concentrations in fish were modelled as an external fixed food source. The calculated concentrations for all these food categories were matched to Belgian consumption data in order to calculate dietary exposure. Estimation of the contribution of deposition in the overall exposure was done by comparing a baseline exposure scenario based on recent atmospheric measurement data with a scenario where dry and wet deposition fluxes were arbitrarily set to zero. Save deposition reference values were derived by comparing the calculated overall exposure for the baseline scenario with the oral Tolerable Daily Intake (TDI). In case of exceedance, the deposition rates were iteratively adjusted until the calculated exposure equaled the TDI. Simulations were ran for 16 priority EPA - PAHs + benzo(j)fluoranthene. For only 4 PAHs sufficient measurement data are available in the EFSA database to perform a reality check. Of these 4 carcinogenic PAHs, only the calculated exposure to benzo(a)pyrene exceeded the TDI with a factor 4, implying that current deposition rates might be too high. More deposition measurement data for BaA/IP are required to confirm these results. (The authors thank the Flemish Agency for Health and Care for their support)

227 A bioassay-directed analysis as a biomonitoring tool to assess the endocrine-disrupting effects of microplastics in freshwater and saline waters

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Air quality is currently assessed by monitoring a few pollutants involved in the pathways of several human diseases. This study aimed to identify with any certainty the molecule responsible for a given biological effect, owing to human co-exposure to many bioactive micropollutants, which can also interact with each other. In this way, in vitro bioassays might be relevant biomonitoring tools to assess the air quality, as they integrate these “cocktail” effects. Furthermore, the pulmonary exposure to semi-volatiles endocrine-disrupting compounds (EDCs) may cause hormonal disruptions observed in humans, especially indoors where they spend 80 % of their time. By using cellular bioassays, we have previously shown that bioactive EDCs tend to concentrate indoors, especially in the gaseous phase. The concomitant chemical analysis of a wide range of identifications of sources in urban areas influenced by particulate emissions from anthropogenic sources. Sampling was performed using Teflon® DA80 high volume samplers between November 2010 and April 2011 in three medium cities located in northern France, Dunkerque (Dk, coastal urban and industrial site), Boulogne-sur-mer (BL, coastal and urban site) and Saint-Omer (StO, inland urban and industrial site). PM2.5 composition was analyzed for major elements, trace elements and major species. Species concentrations were examined according to different ways including temporal evolution, concentration and pollution roses. The impact of such sources on major species and metal concentrations in PM2.5 was then quantified using a weighted non-negative matrix factorization based receptor model that considers constraints on chemical profiles (CW-NMF). NOx, SO2, NH3 and TC were found as the major components of PM2.5 (between 95% and 99%). Only significant differences were evidenced. Trace elements (Cr, Zn, Ni, As, Ag, Cd, Cu, Mn, Pb, V, Sn, Rh, Sr, Bi, Ba, Co, Sb and Tl) only correspond to 0.30% to 0.45% of the PM2.5 mass according to the sites. The CW-NMF model identified 8 common source profiles at the 3 sites: secondary nitrates, residential and road traffic, secondary sulfates, fresh and aged sea-salts, heavy fuel oil combustion, non-exhaust traffic and crustal. In addition, 4 industrial source profiles were identified in Dk and sulphuric emissions and sintering stack of an integrated steelworks, electric steelmaking and glassmaking activities. Despite their low contribution to PM2.5, such industrial sources were the main contributors of metals at the two sites.
of EDCs led to the same conclusions, except in cold season during which the indoor gaseous phase concentrated fewer target EDCs than in summer along with the potential, a bioassay—directed analysis was developed and may represent an advanced biomonitoring tool for air quality. The gaseous phase was collected in a Parisian day nursery during cold season (winter 2014) and in sufficient quantities (6 consecutives samplings of 15 days) to implement the entire fractionation process by semi-preparative liquid chromatography: split of the original extract into three fractions, each fractionated into three subfractions. All these samples and the multiple controls were submitted to the chemical analysis (68 target EDCs) and the biological analysis (estrogenicity measured by transactivation cellular assay). After applying the batteries of target EDCs in the three subfractions (concentration level, intrinsic estrogenicity, lipophilicity), major families found indoors (phthalates, synthetic musks, alkylphenols, parabens) stood out as playing a role in the estrogenicity of the gaseous phase, especially the lead compounds butylbenzylphthalate, galaxolide, butyl-paraben and nonylphenol. The data suggest however the involvement of non-studied EDCs in this biological effect. Our data confirm that bioassays represent suitable biomonitoring tools to assess air quality, whether they are associated with the chemical analysis or not.

228 The Modifying Effects of Ambient Air Pollution on Indoor Air Quality, Impacts on Human Health
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As energy efficient buildings and communities continue to grow, energy retrofits and new designs need to be made in concert with improvements in indoor air quality (IAQ). Although thermal renovations may decrease heat loss and reduce energy expenditures, improved ventilation (airtightness) can affect infiltration and air exchange rates, and magnify the effects and intensity of indoor air pollutants. In this context, a pilot study was conducted to evaluate the tobacco health risk associated with exposure to indoor air pollution in eight buildings ranging from green certified (i.e., LEED Platinum, Living Building) to historic archetypes. The buildings were categorized by the quantity of outdoor air delivered to the in situ ventilation systems, which is situated downwind of major industrial point sources. Indoor and ambient measurements of particulate matter (PM<sub>10</sub>, PM<sub>2.5</sub>), black carbon, ozone (O<sub>3</sub>), total volatile organic compounds (TVOC), carbon monoxide, carbon dioxide (CO<sub>2</sub>), relative humidity, and formaldehyde (HCHO) were collected on a seasonal basis. The heterogeneity in ventilation type along with the negative effects of defective systems and aged mechanical systems had on indoor air quality were distinct; however, the counterintuitive findings implied that green and naturally ventilated buildings underperform when compared to some of the conventional buildings within our study. The CO<sub>2</sub> sensors used in most green buildings respond to the number of occupants within a space but does not consider ambient concentrations of criteria air pollutants (i.e., PM<sub>10</sub>, O<sub>3</sub>, NO<sub>x</sub>) before increasing outdoor air volume. Natural ventilation systems then supply outdoor air to indoor spaces in the absence of mechanical filtration, and in turn compromise the health and well-being of building occupants. Additionally, green and high-performance buildings are equipped with state-of-the-art HVAC systems that work in tandem with occupancy sensors to optimize energy use when buildings are occupied and minimize energy use when buildings are vacant (overnight). To this end, with our limited sample size, our results indicate a correlation between TVOC and HCHO levels overnight. In this given case, the use of occupancy sensors did not allow proper flush-out of indoor environments and interior finishing prior to the building being opened for operations, and as a result, increases exposure over the lifetime of the building.

229 INNOVATIVE AND LOW-COST MONITORING TECHNIQUES FOR EVALUATING THE SPATIAL VARIABILITY OF PM COMPONENTS: VALIDATION AND FIELD APPLICATION
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A new very-low volume sampler has been developed with the purpose of allowing spatially-resolved determination of atmospheric particulate matter (PM) and of its chemical components. The low-cost, automatic and self-powered device assures long-term (1-2 months) collection of PM on membrane filters, suitable for subsequent chemical analysis. The device was deployed during an indoor study focused on the concentration of PM<sub>10</sub> mass, ions, levoglucosane, polycyclic aromatic hydrocarbons (PAH) and elements. It showed very good performance in terms of repeatability of the samplings, which is the essential characteristic to build a reliable network. The samplers have been employed, for the first time, to evaluate the spatial variability of PM<sub>10</sub> mass concentration and its main chemical components in the area of Termini, a urban/industrial hot-spot site in an intramountain depression of Central Italy. Lichen transplants have been exposed at the same sites of the samplers in order to evaluate the potential of lichens as biomonitoring for PM spatially resolved analyses. The meteorological conditions of Terni basin, which limit the dispersion and enhance the accumulation of atmospheric pollutants, are ideal to test and validate new experimental methods for the acquisition of spatially-resolved data.

The added value of using invertebrate species in ecotoxicology: new insights for environmental risk assessment (I)

230 Transgenerational effects of a parental exposure in the sentinel species Gammarus fossarum
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Since the 80s, the development of molecular biomarkers is an important component of ecotoxicology. Unfortunately, field studies that unequivocally link biomarker responses to fitness impacts and finally to population level are scarce (1). This is mainly due to the discordance in time scales between toxicological and ecological responses. In previous laboratory studies exploring the effects of high contamination levels of single molecules, a relationship has been established between genotoxic impacts in gametes of the sentinel species Gammarus fossarum, and impairment in embryo reproduction. However, such a link was not observed after exposure to complex mixtures in the field at more environmentally realistic concentrations (2). Taking advantage of the availability of biomarkers measured in multiple scale in this species, from the molecular level (primary DNA damage, global DNA methylation) to physiological one (feeding rate, molting success, vitellogenesis) and life history traits (growth, fertility, embryonic survival), along with the possibility to conduct rearing culture in the lab (time to puberty about 4 months), the objective of this study was to assess whether biomarker responses recorded in adult gammarids exposed to a chemical stress could be predictive of the fitness of their progeny (i.e. transgenerational effects). For this, the consequences of an exposure in the lab of genitors to environmentally relevant concentrations of cadmium were evaluated in F1 and F2 individuals reared in unremitting conditions. In complement, a field exposure experiment through in situ caging of the adult F0 followed by the assessment of the subsequent effects in F1 and F2 progeny is currently in progress. References (1) Forbes VE, Calow P, Sibbey RM, 2008. The extrapolation problem and how population modeling can help. Environmental Toxicology & Chemistry 27:1987-1994. (2) Lacaze E, Geffard O, Geyot D, Bony S, Devaux A. 2011. Linking genotoxic responses in Gammarus fossarum germ cell with reproduction impairment, using the Comet assay. Environmental Research111:626-634

231 Species differences of bioaccumulation, biotransformation and synergistic effects of two fungicides in two aquatic invertebrates
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Numerous micropollutants have been detected concurrently in aquatic systems, but little is known about the mixture effects of micropollutants in aquatic organisms and the underlying mechanisms. Azole fungicides are known to act synergistically with other chemicals by inhibiting cytochrome P450 (CYP) catalyzed detoxification and thereby enhance the effect of already toxic substances in different organisms. Two widely used fungicide classes co-occur in surface water are strobilurin and azole fungicides. This study aimed to investigate the species' sensitivity to both fungicide classes and to gain mechanistic insights on potential synergistic effects of azoles on strobilurin fungicides in non-target organisms. Therefore, we determined the toxicokinetics of a strobilurin fungicide azoxystrin and an azole fungicide prochloraz in two aquatic invertebrate species Gammarus pulex and Hyalella azteca. Furthermore, we explored median lethal concentrations (LC<sub>50</sub>) of azoxystrin in the presence and absence of prochloraz, the inhibition strength (IC<sub>50</sub>) of prochloraz, and its effect on the locomotory behavior of the two species. Bioaccumulation of azoxystrin were similar in both species with bioaccumulation factors (BAFs) approximately 5 L kg<sup>-1</sup>, while bioaccumulation of prochloraz was different in two species, with BAFs 57 and 110 L kg<sup>-1</sup> in G. pulex and H. azteca, respectively. Many biotransformation products were found for both fungicides in both species of which taurine and malonyl conjugates were only found indoors (phthalates, nonylphenol, polycyclic aromatic hydrocarbons, polycyclic aromatic hydrocarbons, polycyclic aromatic hydrocarbons). Most BTPs result from oxidation and conjugation reactions, which occurred at the (E)-methyl β-methoxyacetyl group of azoxystrin and imidazole ring of prochloraz. Prochloraz inhibited the CYP-catalyzed biotransformation of azoxystrin in both species, leading to higher internal bioaccumulation factors (BAFs) approximately 5 L kg<sup>-1</sup> vs. 1 L kg<sup>-1</sup> in G. pulex.
and *H. azteca*, respectively. The LC50 of azoxystrobin alone were 157 and 200 µg L−1 in *G. pulex* and *H. azteca*, respectively. Prochloraz significantly decreased the LC50 of azoxystrobin in both species. Video-tracking of the locomotory behavior suggested that prochloraz induced hyperactivity in *G. pulex*, but not in *H. azteca.* Overall, results suggests *H. azteca* comprise more diverse biotransformation reactions and *G. pulex* tended to be more sensitive than *H. azteca* toward prochloraz effects.

232 Use of Gammarus sp. for toxicity testing. A case study with the growth regulator insecticide fenoxycarb, H. Araaboune, Istrea Lyon / Freshwater system, Ecology and Pollution Research Unit, Istrea / UR MALY Laboratoire Ecotoxicologie; N. Delorme, K. Abbaci, Istrea Lyon / UR MALY Laboratoire Ecotoxicologie; P. NOURY, Istrea Lyon / Ecotoxicology; R. Tutundjian, Istrea Lyon; E. Vulliet, Institute of Analytical Sciences; G. Daniele, ISA / Biology; C. Barata, CSIC / Environmental Chemistry; I. Fuertes, Institute of Environmental Assessment and Water Research IDAEA CSIC. V. Debat, MNHI / Institute of Systematics, Evolution and Biodiversity

Gammarus sp. (Amphipoda) are widely distributed across European freshwater systems. In the present study, we evaluated the effect of a fenoxycarb exposure on Gammarus sp. More specifically, i) we assessed the sensitivity of the embryo stage, ii) we identified embryogenesis’ sensitive period, iii) we evaluated the sensitivity of the reproductive period and iv) we compared the response to fenoxycarb exposure among three Gammarid species. Fenoxycarb is a growth regulator insecticide, analog of the insect juvenile hormone, used for pest management and for veterinary purpose. This study demonstrated that 5 and 50 µg L−1 fenoxycarb can alter embryonic development of *G. fossarum.* The gastrulation phase was particularly sensitive. Moreover, exposure to 5 and 50 µg L−1 fenoxycarb strongly altered the pre-copulatory behavior in *G. fossarum* and a 50 µg L−1 exposure prevented the production of viable embryos. These results highlighted the deleterious effects of the insect growth regulator fenoxycarb on gammarid embryogenesis and reproduction, which could have severe repercussions on population dynamics. The response to the toxic exposure was dependent on the study gammarid species which underlined the importance to consider species with broader phylogenetic representation to better assess insect growth regulator effects.

233 Adaptation of Gammarus pulex to agricultural insecticide contamination in streams, N. Shahid, Helmholtz Centre for Environmental Research UFZ; J.M. Becker, Helmholtz Centre for Environmental Research UFZ / System-Ecotoxicology; M. Krauss, W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis; M. Liess, UFZ Center for Environmental Research / System-Ecotoxicology

Exposure to pesticides affects non-target aquatic communities, with substantial consequences on ecosystem services. Adaptation of exposed populations may reduce toxic effects of pesticides. However, it is not known whether and under which conditions adaptation occurs when only a low toxic pressure from pesticides is present. Here, we show that Gammarus pulex, a dominant macroinvertebrate species in many agricultural streams, acquires increased tolerance to pesticides when re-colonization from non-contaminated recovery area is low. Populations in the field that were exposed to pesticides at concentrations several orders of magnitude below considerate acute effects showed almost 3-fold higher tolerance to the neonicotinoid insecticide clothianidin (mean EC20= 218 µg L−1) compared with non-exposed populations (mean EC20= 81 µg L−1). This tolerance of exposed populations increased from 2- to 4-fold with increasing distance to the next recovery site (0 to 10 km). We conclude that the development of tolerance for non-target species may occur at very low concentrations, much below those affecting sensitive test organisms and also lower than those predicted to be safe by governmental risk assessment frameworks.

234 The use of antifouling biocides in a changing world: combined impact of nanotechnology and biocides, V.L. Ferreira, University of Aveiro / Biology Department and CESAM; M.D. Pavlaki, University of Aveiro / Department of Biology; M. Monteiro, Aveiro University / Biology; F. Maia, Smallmatt - Small Materials and Technologies, Lda.; R. Martins, Department of Biology, University of Aveiro / Department of Biology and CESAM; J. Tedim, University of Aveiro / Department of Materials and Ceramic Engineering CICECO; A.M. Soares, University of Aveiro / Department of Biology & CESAM; R. Calado, University of Aveiro / CESAM Department of Biology; S. Loureiro, Universidade de Aveiro / Biology

The use of antifouling agents to prevent organism’s adhesion onto surfaces continues to be used worldwide. While the European Union already authorized the use of DCOIT (4,5-Dichloro-2-octyl-2H-isothiazol-3-one) biocide, data on its toxicity to non-target organisms is still scarce. Given the advances in the encapsulation of biocides in smart-releasing systems, this work aims to access the impacts of a new antifouling approach, DCOIT encapsulated in silica nanocounters (SiNC@DCOIT), toward the non-target species Sarcophytom cf. glacium, a coral that is also a model of the cnidarian-algae symbiosis found in some marine invertebrates. Elevated seawater temperatures, as predicted by global climate change scenarios, are described as a major cause of corals reef decline. Due to DCOIT photosynthesis inhibition properties, a joint effect of these two stressors (warmer seawater and DCOIT) may occur in the ocean. Toxicity assays were performed by exposing monocalonial coral fragments (n=5) for seven days, at two different temperatures (present day conditions—26°C—and forecasted scenario for 2100—30.5°C), to 50 µg DCOIT L−1 for free-DCOIT or SiNC@DCOIT and 196 µg SiNC L−1 (nanocontainer control). A negative control was added for each temperature. Photosynthetic parameter (Fv/Fm) was measured using a Pulse Amplitude Modulated Fluorometer (PAM), with the behavioural endpoint (% polyps open) being scored and the biochemical parameters (both in animal and from the fragments) being determined by methods for carbohydrate activity (catalase (CAT), glutathione-S-transferase (GST) and lipid peroxidation (LPO). Results revealed a decrease on Fv/Fm values at 30.5°C, when compared to 26°C (from 0.65 to 0.60), but only in corals exposed to free-DCOIT the temperature effect was significant throughout the days. By the end of the assay the polyps were mainly open at 26°C, whereas at 30.5°C they were closed. Regarding enzymatic activities, significant increase on the GST of both animal and microalgae fractions was found in 30.5°C groups. On the controls, the raise of 4.5°C in water temperature was responsible for a two or three-fold increase in algae and animal GST activity, respectively. The raise on temperature also induced an increment in CAT activity. Regarding LPO, high variability among samples was found. Overall, under thermal stress, the toxicity of DCOIT is enhanced and the negative impacts associated with the use of this biocide will likely be magnified in the warmer oceans.

235 Assessing interspecific variation in Imidacloprid toxicity in earthworms, A. Robinson, Centre for Ecology & Hydrology; E. Lahive, Centre for Ecology and Hydrology; S. Shorth, P. Kille, Cardiff University; D. Spurgeon, Centre for Ecology & Hydrology; A. robinson1,1(alerob@ceh.ac.uk), S. Shorth1, E. Lahive1, P. Kille1, D. Spurgeon1

1 Centre for Ecology and Hydrology, Maclean Building, Benson Lane, Wallingford OX10 8BB, UK 2 School of Biosciences, University of Cardiff, Main Building, Museum Avenue, Cardiff CF10 3AT, UK Widespread interspecific variation is seen in the sensitivity of soil invertebrates to chemical pollutants (e.g. pesticides). Since chemical risk assessments are performed on the basis of tests in a small number of reference species there is a danger of significantly underestimating the effect a toxicant may have in the environment. In order to understand these differences a robust and scientifically based ecotoxicological framework for interspecies ecotoxicological extrapolation is needed. In this study a 30 fold difference in the EC50 reproductive values of imidacloprid was observed across 5 species of earthworms (Eisenia fetida, Lumbricus rubellus, Dendrobaena octaedra, Apporectodea calignosa and Amyntus gracilis) with A. gracilis being the most sensitive and L. rubellus the least. The role of toxicokinetics in determining interspecific variations in sensitivity is interpreted by assessing the Accumulation, Distribution, Metabolism, and Excretion (ADME) of the chemical into the body and to the neurological tissues that are the common target using radiolabelled compounds and cold chemistry. The contribution of toxicodynamic traits to variations in sensitivity was assessed through genome analysis to identify 1) the number, nature and activity of key receptor genes present, and 2) molecular docking affinities as affected by the amino acid substitutions present in different species receptor homologues. Finally, to assess how these interactions affect the key biological and physiological parameters lead to overt toxicity, we use gene expression, biochemistry and life-cycle measurements. By combining these different approaches and identifying key traits, we seek to improve interspecies extrapolation, better predict species vulnerability, and thereby improved the basis for soil species protection during chemical registration.

Solutions for emerging pollutants - Towards a holistic chemical quality status assessment in European freshwater resources (I), M. Santen, G. Ungherese, Greenpeace

Industrial pollution is a severe threat to water resources around the world, particularly in the Global South factories release hazardous chemicals that impact our precious water resources - causing long term devastation to human health and the environment. Rivers supply vital resources, including drinking water, crop irrigation and food. Those who are most critical are the poor. In the past decades Greenpeace did several investigations on persistent and dangerous chemicals (such as PCBs, DCD and DCOIT) in European freshwaters and atmospheres, even identifying a new emerging group of substances, such as pharmaceuticals, personal care products, plastic additives and biocides. Solutions are needed to understand the fate and behaviour of these substances, and to ensure that public health is protected in the future. In this study we outline the issues and developments in the fields of monitoring and assessing the presence of emerging pollutants in freshwater bodies, including detailed site specific monitoring of pharmaceuticals and emerging pollutants in the new River Thames catchment following the Thames Water Incident in 2007.
context of Greenpeace's detox campaign have found a wide range of hazardous substances in the waste waters of textile production or in the effluent of communal wastewater treatment plants (WWTPs) from industrial zones in China, as well as in nearby rivers. Case Studies on per- and polyfluorinated chemicals show that PFAS (perfluorinated alkyl) substances are widespread compounds of environmental concern. Because of their well-recognized hazardous properties, long chain PFASs have been subject to increasing regulation. In 2015 Greenpeace conducted an expectoration in seven rivers, snow and lake water samples were taken at 10 remote high altitude sites showing that these persistent chemicals are present everywhere on the planet. In 2017 Greenpeace Italy carried out PFAS analysis in wastewaters, analysis revealed PFASs presence in all tested samples of rivers and drinking water collected in schools and public fountain. It is not too late to act – but new rules and regulatory measures are required. The use of pollution prevention control or wastewater treatment does not deal effectively with all hazardous substances, and only postpones the need for more effective measures. The problem has to be tackled at its source. The Detox campaign challenges top textile brands to work with their suppliers and eliminate PFAS and all other hazardous chemicals across their entire supply chain, and the entire life-cycle of their products. The growing concern about Europe’s massive pesticide use goes hand in hand with an increasing need to search for ecological solutions. To be effective, action needs to be based on knowledge, which requires transparency as a first step, the quantities of hazardous substances used and discharged to be reported and monitored, with full availability of data to the public. [1] 

B. Escher, Helmholtz-Centre for Environmental Research GmbH - UFZ / Cell Toxicology; F.D. Leusch, Griffith University / Australian Rivers Institute; P.A. Neale, Griffith University / School of Environment; A. van Wezel, KWR Watercycle Research Institute / Chemical Water Quality and Health; M. Dingemans, KWR Watercycle Research Institute; M. Meeker, Water Environment & Reuse Foundation (WE&RF) Bioanalytical tools hold great promise as an additional tool of our current water monitoring strategies. In vitro bioassays, which are increasingly being applied in water quality assessment, provide relevant and robust predictive biosystems able to specifically and quantitatively measure early adverse effects of contaminants in water, including providing a measure of mixture effect, even in low doses, where included compounds alone are not measurable. One approach along these lines include comprehensive and high-throughput monitoring systems for a wide range of water contaminants, without the use of experimental animals. Smart combinations of chemical & biological analytics can lead to reduced uncertainty in safety assessments, especially with regards to endocrine disruption, oxidative stress as other relevant primary adverse outcome pathways triggered by environmental mixtures of water contaminants. Gathering the experts worldwide, recent large scale projects delivered several methodological advances leading to a comprehensive framework including the most promising panel of assays and expanded effect-based trigger values (EBT) for both drinking water and environmental waters (GWRC Endocrine Toolbox II, FP7 DEMENTAU, FP7 SENSITIVE, BRATON), thereby making this innovative approach contribute to strengthen the safety of conventional water treatment plants and be integrated in future regulations. They also could provide robust monitoring frameworks to promote alternative water schemes as promoted by the Blue Print Initiative in Europe to better safeguard water resources and the WHO Potable Reuse Guidance document. While leading players in Australia, Europe and US recommend to incorporate proactive tools for risk assessment of PFAS Australia, US (CA), Canada, RIVM, EAWAG, KWR, UFZ, EU-JRC and EU DG-Env, WHO and GWRC), these bioanalytical tools need to be more comprehensively validated and benchmarked across the entire water cycle and against human and ecological health outcomes before they can be adopted in regulatory frameworks. A critical next step will be to derive further EBT for an expanded scope of bioassay endpoints. Several strategies for the derivation of EBT have been proposed but there remains a lack of acceptance and harmonization across the field to allow better acceptance of these innovative water quality and safety frameworks. Covering a wide range of issues including water quality and quantity management and the management of water-related risks, the OECD is endeavouring to capture science as policy recommendations that derive from its past and recent work on water in a single, consistent and action-oriented policy. By hosting a collaborative task-force or expert working group including GWRC experts and gathering international organizations such as WHO, UNESCO and the OECD we can get to benchmark these new effect-based trigger values, and contribute to the water challenge by targeting Water effect-based guidelines. Complementary tasks could be opened for the assessment of mixture effect to an action Science to Policy interface as a supportive action to better explain and disseminate the associated benefits for stakeholders as citizen towards their health protection, municipalities and local authorities, water professionals and institutional bodies. 238 Chemicals of emerging concern (CEC) in the water cycle – a regulatory perspective M. Helmecke, Umweltbundesamt (UBA) Environmental authorities increasingly need to address the challenge of contaminants of emerging concern found in the water cycle. The German Environment Agency has assessed entry points, critical characteristics of chemicals and the existing legislation to derive potential measures to minimize micro-pollutants in the aquatic environment. A holistic and precautionary approach is needed that combines measures at the source, during the usage of products and chemicals as well as end-of-pipe measures. The EU Water Framework Directive and the Marine Strategy Framework Directive pose a legal frame to achieve good status of all waterbodies and prohibit any further deterioration. Environmental waters (GWRC Endocrine Toolbox II, FP7 DEMEAU, FP7 DEMENTAU) expanded effect and comprehensive framework including the most promising panel of assays and mixtures of wate. Bioanalytical tools hold great promise as an additional tool of our current water monitoring strategies for a wide range of water contaminants. Complementary tasks could also be taken up by such Science to Policy interface as opportunity in this regard as agreed by the European Water Directors in 2016. However, there are challenges regarding the inclusion of new approaches to a regulatory context. 239 Non-target Screening for Holistic Chemical Monitoring and Compound Discovery: Open Science, Real-time and Retroactive Approaches E. Schymanski, University of Luxembourg / Luxembourg Centre for Systems Biomedicine (LCSB); R. Alizadeh, National and Kapodistrian University of Athens / Department of Chemistry; N. Alygiakis, Environmental Institute; J. Hollender, Eawag / Environmental Chemistry; M. Krauss, T. Schulze, Helmholtz centre for environmental research - UFZ / Effect-Directed Analysis; J. Slobodnik, Environmental Institute; N.S. Thomaidis, National and Kapodistrian University of Athens / Department of Chemistry; A.J. Williams, US EPA / ORDNCCT Non-target screening (NTS) with high resolution mass spectrometry (HR-MS) provides new opportunities to discover new contaminants, their dynamics and effects on the environment far beyond the current 45 “priority pollutants” or even “known” chemicals. Open science and the exchange of information (between for example scientists and regulatory authorities) has a critical role to play in the continuing evolution of NTS. Using a variety of case studies from Europe, this talk will highlight how open science activities such as MassBank.EU (https://massbank.eu), the American Mass Spec Monitor (http://msmonitor.jpl.nasa.gov) and NORMAN Sample Freezing Platform (http://norman-data.eu) as well as the US EPA CompTox Chemistry Dashboard (https://comptox.epa.gov/dashboard) can support NTS. Further, it will show how initiatives such as near “real time” monitoring of the River Rhine and retrospective screening via so-called “digital freezing” platforms have opened up new potential for exploring the dynamics and distribution even of as-yet-unknown chemicals. Collaborative European and international activities facilitate data exchange amongst analytical data scientists and enable quick, effective and reproducible provisional compound identification in digitally archived HR-MS data. This is leading to new ways of assessing and prioritizing the new generation of “emerging pollutants” in the environment, enabling a pro-active approach to environmental challenges that we cannot expect to be minimized within a foreseeable future. This abstract does not reflect US EPA policy. 240 Toxicological profiling of water samples with in vitro bioassays and assessment using effects-based trigger values B. Escher, Helmholtz-Centre for Environmental Research GmbH - UFZ / Cell Toxicology; R. Altenburger, UFZ Centre for Environmental Research / Department Bioanalytical Ecotoxicology; S. Alt-Atis, Institut National de l'Environnement Industriel et des Risques (INERIS); P.A. Behnisch, Biodetection Systems BV; W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis; F. Brion, INERIS / Ecotoxicology Unit; S. Buchinger, Federal Institute of Hydrology / Department G Biochemistry Ecotoxicology; W. Busch, Helmholtz Centre for Environmental Research - UFZ GmbH / Bioanalytical Ecotoxicology; S.E. Crawford, RWTH Aachen University / Institute for Environmental Research,
with adverse outcome pathway knowledge, can guide research observed biological responses using data from fathead minnows exposed to site contaminants. More specifically, we use chemical effects data to evaluate risks associated with chemicals present in the environment. Evaluating the potential human health and ecological risks associated with exposures to complex chemical mixtures in the environment is one of the main challenges of chemical safety assessment and environmental protection. There is a need for approaches capable of integrating chemical monitoring and biological effects data to evaluate chemicals present in the environment. We will present an approach that uses prior knowledge regarding the biological effects of the effluent of a local wastewater treatment plant (WWTP) to develop chemical lists of EQS, the proposed generic methods to derive EBTs is a first step to develop a method that reads across from existing EQS and makes additional mixture considerations to assure that the derived EBT are protective for complex mixtures as they occur in surface water. The EBT derivation method was applied to 48 in vitro biosassays with 37 of them having sufficient information to yield preliminary EBTs. 30 of those were considered robust enough to pursue further and for the remainder it is necessary to obtain more experimental data for single chemicals but also to derive more EQS values. To assess the practicability and robustness of the proposed approach, we tested the EBTs numerous case studies from the literature where wastewater treatment plants and surface water were evaluated with biochemical tools. In this presentation, we highlight specifically case studies from the EU project SOLUTIONS, where water quality was assessed in large streams (e.g., Danube), hot spots of contamination (e.g., disposal of uncontrolled wastewater in the river Rhine tributary Bregenzer Aare), and in viral treatment plant effluent into small creeks (case study of small Rhine tributaries in Switzerland). In many cases the proposed EBTs were able to differentiate wastewater from surface water and EBTs for different biosassays gave very consistent results indicating the benefit of a common derivation method. Despite the limitations due to limited effect data availability and limitations of the existing lists of EQS, the proposed generic methods to derive EBTs is a first step to harmonise existing approaches and explore various different options of a large diversity of in vitro biosassays commonly applied for water quality assessment.

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241 Chemical gene interactions for associating contaminants with biological effects

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Evaluating the potential human health and ecological risks associated with exposures to complex chemical mixtures in the environment is one of the main challenges of chemical safety assessment and environmental protection. There is a need for approaches capable of integrating chemical monitoring and biological effects data to evaluate chemicals present in the environment. We will present an approach that uses prior knowledge regarding the biological effects of individual contaminants to predict toxicity of mixtures and prioritize contaminants. More specifically, we use chemical-gene interactions networks to develop knowledge assembly models (KAMs; which is specific to the aquatic system of interest) based on chemical monitoring data and publically available chemical-gene interaction data. When only chemical data are available, KAMs allow for the development of site-specific hypotheses for follow-up biological effects testing. When transcriptomics data are available, KAMs can be used with statistical approaches, such as reverse causal reasoning approaches to prioritize risk and contaminants. Two brief examples using chemical-gene interactions and KAMs will be presented. The first example used chemical monitoring data from the effluent of a local wastewater treatment plant (WWTP) to develop chemical-gene interaction networks. The networks were used to develop hypotheses about the biological effects of the effluent. To test the network predictions, targeted gene expression, using quantitative polymerase chain reaction, was measured from adult male and female fathead minnows that were exposed to the effluent. The second example presented preliminary predictions about chemical-gene interactions to develop a KAM for detected chemicals at five locations near two WWTPs. Hepatic transcriptomic data from fathead minnows exposed to site-water at each location were mapped to the assembly models to evaluate the likelihood of a chemical contributing to the observed biological responses using richness and concordance statistics. The use of chemical-gene interaction networks and KAMs have strong potential for associating chemical occurrence data to biological effects that, when integrated with adverse outcome pathway knowledge, can guide research and/or monitoring efforts related to the effects of contaminants in the environment. The contents of this abstract neither constitute nor necessarily represent official US EPA views and policies.

Anthropogenic and natural sources of environmental contaminants highlight the impacts of opposing and conflicting regulations

242 The triazole story: Clarification of sources, fate and footprint in the environment of the molecule 1,2,4-triazole

M. Blank, Bayer AG Crop Science Division / Environmental Safety; H. Borchers, A. Changelle, Bayer Crop Science AG; G. Telschek, Bayer AG Division, CropScience/Environmental Fate / Development Environmental Safety; O. Heinemann, Bayer Crop Science Division

The triazole 1,2,4-triazole (124T) is a key structural component of azole fungicides, one of the world’s most widely used fungicide classes in agriculture. Crop protection industry taskforce (Triazole Derivative Metabolite Group, TDMG) is jointly addressing scientific and regulatory topics, covering the environmental fate, metabolism, ecotoxicology, toxicology, and risk assessment of 124T. So far in the regulation of crop protection its origin was interpreted as a result of the breakdown of the parent azole-fungicides only. 124T is toxicologically classified as R63. According to the EU Regulation, Directive 11/2009, it is a ‘relevant’ metabolite in groundwater and subject to a legal maximum concentration of 0.1 µg/l with respect to crop protection uses. Distinct restrictions on agricultural uses of azole-fungicides have been necessary to meet this hazard based limit value. In response to the large number and widespread use in the EU of registered azole products, regulatory authorities are asking for information about 124T’s potential leaching and actual concentrations in groundwater. The TDMG scientists discovered that 124T occurs ubiquitously in the environment, originating also from other anthropogenic sources than azole-fungicides, such as fertilizer additives (to avoid nitrate leaching) and commodity chemicals, and is also naturally formed. To investigate the different sources of 124T and elucidate the pathways of entry and distribution in the environment innovative approaches were needed. The TDMG scientists have therefore applied omics (and new scientific scope into non-agricultural environments and residues in different matrices). The work is supported with a recent terrestrial field dissipation study using a 13C stable isotope labelled azole-fungicide to enable differentiation between the different sources, a forest soil study to assess the background abundance of 124T in non-agricultural soils, and several groundwater monitoring studies. This work has confirmed the wide range of sources of 124T and showed that it is currently not possible to distinguish between their contributions to measured environmental concentrations. Consequently, relying on azole-fungicides as the only source of 124T-concentrations could result in a systematic over-estimation of the environmental exposure risk from pesticide usage. An improved understanding of the sources of the molecule in the environment is a prerequisite for reliable and justified regulatory conclusions.

243 The triazole story: Assessment of the background abundance of 1H-1,2,4-triazole in selected German forest soils

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1H-1,2,4-triazole (124T) is an ubiquitous small molecule which originates from different anthropogenic sources in the environment or from biotic or abiotic degradation of triazole-fungicides. In addition, 124T potentially originates from natural sources like soil microorganisms (Fungi, Actinomycetes). Information about the natural background abundance of 124T in forest top soils of German origin is of importance for the assessment of the entry paths and occurrence levels into the environment by crop protection measures. In a GLP terrestrial field study, duplicate forest top soil samples from ten different locations and different forest types (beech, spruce, pine, oak) in Germany were sampled in 2012/2013 for analytical investigation of the 124T background. For this reason, remote areas without close contacts to agricultural areas were selected. In addition, at two of these sites the development of the 124T residue background level was investigated over the period of one year. The background abundance of 124T in the samples ranged from < 1.0 to 1.9 µg/kg in oak forest top soils, from 1.0 to 2.1 µg/kg in pine forest top soils, and from < 1.0 to 1.2 µg/kg in spruce forest top soils. In the selected beech forest top soils the background abundance of 124T was below 1.0 µg/kg. The background abundance of 124T in beech and spruce top soil samples taken from April 2012 to February 2013 showed fluctuations over time. These variations could not be associated to seasonal changes. Single values ranged from < 1.0 to 1.8 µg/kg in the beech top soil samples and from < 1.0 to 2.1 µg/kg in the spruce top soil samples. Overall, a background abundance of 124T could be determined in all forest top soils. The evaluation clearly showed that anthropogenic 124T sources (e.g. fertilizer additives, metabolites of triazole-fungicides, breakdown product of commodity chemicals) could be excluded. This indicates that the measured 124T residues originate from natural sources in the environment.
Challenges of a groundwater monitoring study design for a substance with multiple sources: determining risk for groundwater from 1,2,4-Triazole formed from fungicides used in arable crops in Germany

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1H-1,2,4-Triazole (124T) is a widely occurring molecule with a number of anthropogenic sources, but also biogenic sources, all of which may result in the presence of the molecule in groundwater. The regulatory framework under which potential risks to environment and health are assessed, including applicable trigger concentrations, depends on the source. As a metabolite of triazole fungicides used in agriculture, 124T is regulated under EU Regulation 1107/2009 and subject to a regulatory trigger concentration of 0.1 µg/L in groundwater. At the same time, 124T is also used in agriculture as a nitrification inhibitor added to mineral fertiliser or slurry, making this a potential additional source contributing to overall soil load. The risk to groundwater from this use is however assessed under other regulations.

To address regulatory concerns about the leaching risk for 124T from combinations of triazole fungicides used in agricultural practise, the industry group TDMG has conducted groundwater monitoring studies in Germany. As the aim was to evaluate the risk from triazole fungicides only, the challenge was to design studies to sample groundwater originating in areas with intensive triazole fungicide usage, but where applications of 124T-containing fertiliser and other potential sources could be reasonably ruled out. Existing wells from authorities’ or water producer’s monitoring networks were sampled in the studies, thus capturing a range of scenarios for leaching risk in real-world agricultural practice. To rule out other anthropogenic sources of 124T, for each potential monitoring well a stepwise screening approach was applied to ensure suitability. This, with in-depth farmer interviews to document relevant product applications and rule out use of 124T-containing fertilizer in the upstream infiltration area. In total 211 groundwater samples from 31 wells in different regions of Germany were analysed. All samples were < 0.1 µg/L, with 14 samples between 0.05 (=LOQ) and 0.08 µg/L. The results show that even with extensive use of triazole fungicides, the concentrations of 124T in shallow groundwater downstream from treated fields did not exceed the regulatory trigger of 0.1 µg/L. The presented approach is considered to be effective to obtain a realistic picture of groundwater exposure to 124T from triazole fungicides in agricultural practice. Ruling out other sources is challenging, but possible with site screening and engagement with the farmers.

247 Overlooked sources of trifluoroacetate in the water cycle - consequences for drinking water supply and regulatory measures

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Relevant amounts of trifluoroacetic acid (TFAA) are formed in the atmosphere by photochemical transformation of several refrigerants and subsequently introduced into the aquatic environment by wet deposition. TFAA occurs as trifluoroacetate (TFA) in the aquatic environment and is considered to be persistent and mobile. Both the acid and its potassium salt are manufactured and/or imported in large amounts in the European Union. Most of the TFA produced in the world is used in the biodegradation product of several pesticides. During a screening of surface waters in south-west Germany, high concentrations of TFA (up to 140 µg/L in a tributary of the River Rhine) were detected. As a consequence, concentrations of TFA at adjacent bank filtration sites and tap waters were also substantially elevated. The here presented study aims on source identification as well as on the assessment of treatment options (ozonation, chlorination, activated carbon filtration) for contaminated raw waters. Ozonation of model substances and wastewater samples was applied to reveal the TFA-forming potential of individual compounds and the presence of not yet identified TFA-precursors in waters bodies in general. Discharge of industrial wastewater was identified as the source of elevated concentrations of TFA in the tributary of the Rhine river. Extended monitoring demonstrated that this contaminant still impairs the drinking water supply along the lower River Rhine. Ozonation, activated carbon filtration, and chlorination do not allow for considerable removal of TFA. Monitoring of wastewater treatment plants (WWTP) demonstrated that WWTP also emit TFA. One WWTP showed substantial formation of TFA and ozonation of WWTP-outflows led to increasing concentrations. Tests on the formation of TFA by ozonation of the selected potential precursors demonstrated highly compound-specific TFA-yields between insignificant and ~40%. Sources and pathways of TFA may still hold hidden. As contaminated water resources cannot be efficiently treated by technical options typically applied in water utilities, this topic needs to be addressed for sustainable raw water management. Small molecules such as TFA may originate from different sources and their individual contributions may be subject of temporal and spatial variability. This must be taken into account with regard to risk assessment as well as for the regulation and authorization of chemicals.

Persistence & Biodegradation Assessment

248 Why biodegradable chemicals persist in the environment? A look at bioavailability

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The biodegradability of a given chemical in the in the environment cannot be assessed properly without considering the chemical’s bioavailability to the degrading microbial populations. This is especially applicable to hydrophobic organic chemicals (HOCs), like polycyclic aromatic hydrocarbons (PAHs). With the aim of providing pathways for implementation into regulatory contexts, this...
sensitive to temperature fluctuations. Our study highlights limitations in the Arrhenius model predictions in the 0°C to 40°C range. Consequently, model estimations could not accurately predict the biotransformation potential at higher temperatures, where the microbial community was also monitored. This study was conducted at 20°C. The geometry of the container resulted in a water depth of 140 cm, and a surface area of 0.70 m². In contrast to OECD 309, the system is exposed to direct photolysis in outdoor water bodies by the sampling procedure. A second container with the same test setup but equipped with a lid of stainless steel served as a control. The test is conducted as both pelagic and suspended-sediment setup. Test substance was 14C-pentadichlorophenol, which is known to degrade rapidly in aquatic systems under the influence of light. The purpose of the test was to determine if photolysis is a relevant process in natural water bodies under OECD 309 test conditions and up to which water depth indirect photolysis might contribute to degradation since the light intensity decreases with increasing water depth. A simulation approach has been performed considering the major conditions required in OECD 309 but at a much larger scale. Stainless steel containers of 900 liter volume are filled with surface water taken from a natural lake maintained at 20°C. The geometry of the container was 140 cm and a surface area of 0.70 m². In contrast to OECD 309, the system is exposed to simulated sunlight and the water is not mixed by stirring or shaking. Sampling is performed in different water depths using permanently installed steel tubes of different length in order to avoid mechanical mixing of the water body by the sampling procedure. A second container with the same test setup but equipped with a lid of stainless steel served as a control. The test is conducted as both pelagic and suspended-sediment setup. Test substance was 14C-pentadichlorophenol, which is known to degrade rapidly in aquatic systems under the influence of light. The purpose of the test was to determine if photolysis is a relevant process in natural water bodies under OECD 309 test conditions and up to which water depth this can be applied.

251 Findings from an international ring test for an improved marine biodegradation screening test

A series of standardised biodegradation screening tests (BSTs; e.g. OECD 301, 306) have been developed to measure the relative biodegradability of chemicals. Recently, regulatory emphasis has shifted from measuring biodegradation towards prioritisation on chemical persistence. In their current guise, BSTs are ineffective as screens for persistence. They are prone to high levels of variation and produce a large number of fails, many of which can be considered false negatives, whereby a chemical fails a test not because of its recalcitrance, but rather because the test itself has failed. An ECETOC funded workshop to discuss improvements to marine biodegradation testing was delivered in 2015. During this workshop, methodological improvements to BSTs were discussed, in addition to clarifying guidance on testing and interpretation of results obtained from marine BSTs. Methodologically: (i) increasing bacterial cell concentrations to better represent the bacterial diversity inherent in the sampled environments; and (ii) increasing test durations to investigate extended lag phases observed in marine assessments, were recommended to be validated in a multi-institutional ring test. This presentation will report the findings from an international ring test of an improved marine BST, whereby an improved marine BST comprising inocula concentrated by tangential flow filtration, a modified marine BST comprising seawater and a standard OECD 306 closed bottle test were compared across 13 laboratories in the UK, Norway, Germany, Italy, Canada, USA and Japan. Five test chemicals including a positive reference compound (sodium benzoate), a negative reference compound (pentaehlorophenol) and three compounds with variable reported degradation (4-nitrophenol, triethanolamine and hydrolysed polycrylamide) were used. The test was performed to batch measure biodegradation at defined conditions. Other processes which might be relevant for the fate of a chemical in marine biodegradation assessments. The role which microbial communities play in chemical biodegradation and the extent to which microbial community analysis can explain intra- and inter-laboratory variation in biodegradation test outcome will also be discussed.

252 Relevance of photolysis for the fate of pentadichlorophenol in deeper water layers - results of a scale-up approach according to OECD TG 309

D. Hemmcke, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Ecological chemistry; M. Kruse, Fraunhofer IME, Institute for Molecular Biology and Applied Ecology; J. Hassink, BASF SE / Environmental Fate, OECD TG 309 "Aerobic Mineralisation in Surface Water" is currently used under different regulatory frameworks for the persistence assessment of chemicals in surface water. The test is performed in batch to measure biodegradation at defined conditions. Other processes which might be relevant for the fate of a chemical in the aquatic environment are oxidative processes, like direct and indirect photolysis, and hydrolysis. This study investigated the relevance of the different substances and their impact on the test results. It was found that for pentadichlorophenol and three compounds with variable reported degradation (4-nitrophenol, triethanolamine and hydrolysed polycrylamide), were used. The test was performed to batch measure biodegradation at defined conditions. Other processes which might be relevant for the fate of a chemical in marine biodegradation assessments. The role which microbial communities play in chemical biodegradation and the extent to which microbial community analysis can explain intra- and inter-laboratory variation in biodegradation test outcome will also be discussed.
Integrating life cycle approaches towards a sustainable circular economy (I)

254 How can we measure a sustainable circular economy? Unveiling current indicators for the life cycle of products

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Currently, EU policy on circular economy (CE) aims to decrease environmental damage as well as secure the future supply of resources to support economic growth. Even though the implementation of new strategies might cause burden shifting, it is mostly assumed that an increased circularity of resources results in environmental benefits. At the same time, indicators suggested to assess CE progress often fail to provide an assessment of both CE goals and strategies from a sustainability perspective. A life cycle perspective provides a point of departure to address CE strategies, as the stages involved in the circulation of materials are clearly illustrated. Nevertheless, which indicators to assess is still to be defined to support the implementation of CE at any stage of the supply chain. This contribution aims to identify the type of indicators suggested to measure the progress towards a CE at a product level and to evaluate these in relation to the overarching goals and the implementation strategies of CE. To this end, we first define the main CE goals and implementation strategies identified in recent literature and translate these into measurable flows by creating a system model that accounts for each step in the product life cycle. Finally, we review the literature on CE indicators and classify them into CE goals and strategies, life cycle stages and flows addressed, and measurement units (i.e. economic, mass, energy or environmental impact). This contribution provides a consistent framework to compare and assess CE performance indicators at a product level. It also aims to develop a companion decision analysis framework that supports CE. Through a structural assessment of currently suggested CE performance indicators, we can define gaps and needs in the monitoring process of CE to ensure that progress contributes to the overarching goals of CE and in turn capture trade-offs between implementation strategies.

255 Making sense of circularity indicators with Multi Criteria Decision Analysis

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The focus of this study is on packaging, i.e. a sector with high priority for circular economy (CE) implementation, by exploring a situation where a company intends to compare the circularity performances of different products in order to identify which is the best option from a CE perspective. We considered six different packaging alternatives for beer in different contexts and calculated the following indicators to assess product-level circularity: i) the Material Reutilization Score (MRS), included in the Cradle to Cradle® certification program; ii) the Material Circularity Indicator (MCI) developed by the Ellen Mac Arthur Foundation and Granta and iii) the most relevant impact categories according to the Product Environmental Footprint Category Rules for beer product category, i.e. climate change (CC), acidification (Ac), and marine eutrophication (ME). The MCI and TOPSIS (Technique for Order of Preference by Similarity to Ideal Solution) are integrated through an integration approach to aggregate indicators representing performance of the product system with respect to various aspects such as material recyclability, recycled content and eco-efficiency. We argue that ranking the alternatives based on such approach will ease the identification of the best packaging alternative from a CE perspective. The MCDA is applied to process the indicator scores and subsequently obtain the ranking. Specifically, the compensatory approach based on the MCDA method TOPSIS (Technique for Order of Preference by Similarity to Ideal Solution) is used for ranking the six alternatives. The results of the stand-alone application of the two types of indicator sets, i.e. those focusing only on circularity (MRS and MCI) and on life cycle assessment (CC, Ac, ME) are different. The ranking of the packaging is the same within the life cycle assessment indicators, but differs when MCI and MRS are considered. The implementation of the MCDA with different weighting sets shows that two alternatives are dominating i.e. have higher scores for all the indicators. These two alternatives are ranked in the first two positions in all the weighting schemes and hence the ranking is considered as stable. The use of MCDA in combination with several product-circularity indicators is thus recommended to support companies in the identification of the best alternative from a CE perspective.

256 Consistent allocation using archetypes of LCA Goal and Scope definitions

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Identifying a suitable allocation procedure is always a challenge in the modelling of the life cycle. This is especially the case for metals [1]. However, by recommending both partitioning and substitution for establishing the data for an attributional LCA, several inconsistencies are introduced. It becomes unclear to what research question LCA results respond if different modelling approaches are combined in a single LCA study. We developed archetypes of LCA Goal and Scope definitions – in the form of research questions – that aid in identifying a suitable and consistent allocation procedure. In order to identify a consistent LCA study, we performed LCA implementations of the LCA goal and scope need to be clearly defined: the topic of the LCA, the perspective, the reason to conduct the study, and potential additional functions of the product system that are taken into consideration. We present a framework that shows how the allocation procedure is dependent on the different LCA approaches that are defined in the goal and scope of the LCA. Based on this framework, building blocks are derived that are used to formulate research questions. These research questions represent archetypes of goal and scope definitions. The presented framework shows that there is a relevant difference between system expansion and substitution, and that we must differentiate between process-oriented and product-oriented LCAs, which is not common practice. Furthermore, we show that all types of LCA approaches can be used to support decision-making, which is often only ascribed to consequential LCAs. It is concluded that it is not the topic but the research question of the LCA study that determines the most suitable allocation procedure. One LCA topic (e.g. 1 kg of recycled aluminium) can already be used for at least 13 different research questions. “What is the impact of 1 kg of recycled aluminium?” is not detailed enough to identify an LCA modelling approach. This paper shows the level of detail that is required to enable this. The importance of a research question is not always acknowledged in LCA practice, but is crucial for unambiguous interpretation and communication of LCA results. [1] PE International, Harmonization of LCA Methodologies for Metals, Ottawa, Canada, 2014.

257 Sustainability assessment of product lifetime extension through increased repair and reuse

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The concept of circular economy is characterized by an economy that aims to keep products, components and materials at their highest utility and value for as long as possible. Based on data collected by a reuse organisation, computers are often considered for self-repair by consumers. In order to increase the number of successful repairs and reduce the required time, current European policy aims to improve the repairability of products. The potential environmental benefits of reuse after repair is investigated by considering a baseline, recycling and reuse scenario. The baseline scenario is considered to be the worst case because it does not consider any recycling or reuse and it assumes all waste is incinerated. A professional use of 3 years is assumed with an annual electricity usage of 76 kWh from the average European grid. For disposal, a specific incineration dataset was calculated based on the assumed laptop computer using the available ecoinvent waste tools. In the recycling scenario the laptops are collected and sent to a recycling plant for material recovery. The result is a closed loop for recovered materials. For this reason, only a limited number of materials are taken into account. The recycling scenario assumes the optimistic case that all laptops are collected. The pre-processing of the laptop is modelled assuming a manual depollution step followed by mechanical treatment. The end-processing is modelled assuming remelting in an average European electro furnace, aluminium production site and cement kiln as material recycling technologies. In the third scenario, a second life of 3 years is assumed for the repaired laptop. The main environmental impact of the repair activity is the replacement of components. It is assumed the laptop requires a new hard disk drive and a new battery. No additional transportation is assumed for self-repair. At end of life the laptop is assumed to be recycled, as described above. The case study presented in this paper indicates that repair of laptop’s should be considered before discarding for material recycling. The potential benefits of material recycling for high-end or closed loop applications remain limited. If the extended life is at least 2 years, the number of components to be replaced considered in this case study does not impact the decision making. The results also show that increased energy efficiency of new products has a limited impact on the overall results.

258 Building - Rooftop Symbiosis at the next level. Improving urban agriculture through circular economy strategies

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Cities food supply has become an emerging problem with consequences like greenhouse gas emissions or land occupation. In this direction, new ways of producing food within the limits of cities have arisen as potential solutions. Integrated Rooftop Greenhouses (i-RGT) have the potential of a conventional greenhouse for producing vegetables, but they are located at the top of a building,
where they can benefit from different residual flows that were previously being wasted. In ICTA-ICP building (4 floors), in the UAB campus (Bellaterra, Barcelona), a rainwater harvesting system stores rainwater on an underground tank, from which water is pumped to the rooftop to irrigate the crops with a hydroponic automated system. In the third floor, high CO2 concentrations (up to 820ppm) and more stabilized temperatures (between 15 and 22ºC) are reached. The transport of the soil to the rooftop could benefit crop production by performing a CO2 enrichment and providing more suitable temperatures to the plants. In this aspect, an open chamber made of steel and LDPE will be used to compare its environmental performance and production with a control crop, using life cycle tools. In this direction, previous LCA studies have stated that the fertilizers are one of the items that exerts the greatest impacts in i-RTG systems. Hence, different nutrients need to be optimized. In this sense, different literature expresses that half of the currently economically phosphate resources will have been used up in 50 to 100 years. To optimize P cycle, struvite has been defined as a potential source of this nutrient. With a circular economy perspective, wastewater treatment plant precipitated struvite will be used as the P source for crops in the i-RTG in two different ways: mixing struvite in the nutrient solution and by adding struvite in perlite sacks (hydroponic substrate). The goal of this contribution is thus to evaluate the CO2 and P cycles in an i-RTG through experimental and environmental studies by considering circular economy strategies. With these enhancements, urban agriculture will cut its environmental impacts, making it a more sustainable source of food for cities.

259 Chemical recycling of plastic packaging waste - A life cycle perspective on PET recycling

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Plastic packaging waste is one of the priority areas inside the European action plan for the transition to a circular economy. To establish a circular economy, a potential large-scale avenue is chemical recycling of plastic packaging waste. However, preliminary assessments of chemical recycling technologies rate them inferior compared to mechanical recycling not only from an economic but also from an environmental point of view. Based on these results, we develop a method based on life cycle assessment to evaluate the environmental performance of chemical recycling to produce chemicals. The method calculates the maximal environmental impact reduction for chemical recycling technologies for future industry setups. To calculate the maximal environmental reduction, a general model of the life cycle of plastic packaging waste is developed from cradle-to-grave. The results are benchmarked against both mechanical recycling and energy recovery for the environmental impact categories climate change and fossil resource depletion. The basis of the calculations are four key system parameters: (i) the efficiency of the waste treatment technology and the environmental impacts of (ii) production of secondary materials, (iii) the waste treatment technology and (iv) the target chemical produced by chemical recycling. The presented method is used to evaluate chemical recycling technologies for polyethylene terephthalate (PET). In this case study, we show the need to produce high value-added chemicals from chemical recycling technologies to possibly achieve an environmental benefit compared to mechanical recycling. To improve on mechanical recycling, chemical recycling needs to yield a chemical that is currently produced with an impact on climate change or fossil resource depletion exceeding 2.54 kg CO2 eq. or 1.58 oil eq. per kg, respectively. To the best of the authors’ knowledge, no chemical in patents or literature meets both of these threshold values. In contrast, chemical recycling is beneficial compared to energy recovery in all studied cases. The presented method enables the easy and early-stage assessment of the maximal environmental reduction of chemical recycling. The case study shows that chemical recycling should target PET waste that is currently used for energy recovery or needs to transform waste from mechanical recycling to high value-added chemicals.

Informed substitution of hazardous chemicals for circular economy: science and practice

260 Substitution of PFOS under the Stockholm Convention


In 2009 PFOS, its salts and PFOSF were added to the UNEP Stockholm Convention aiming at protecting human health and the environment from POPs. However, the addition contained twenty exempted uses, for which PFOS could still be produced and used. These exemptions were accepted, accompanied by a decision to evaluate uses at regular intervals in order to facilitate a total phase out. Besides the evaluation, the Convention provides Guidance of alternatives to PFOS, which is regularly updated and meant to facilitate the Parties to the Convention in phasing out PFOS. The evaluation was carried out in 2014 under the POPs Review Committee. In 2016 the previous Guidance on alternatives was updated, followed by endorsement in 2017. The update focused on all known applications of PFOS, including the twenty exemptions. However, priority was given to the open applications in two pesticide uses, namely insect baits for control of leaf-cutting ants from Atta spp. and Acromyrmex spp. and insecticides for control of red imported fire ants and termites. Data on these applications update will be presented. The presentation will focus on the further need of PFOS and the specific uses in various Parties under the convention and possible alternatives among which chemical and non-chemical ones. The recommendations of the POPs Review Committee led to the decision to request the Parties using PFOS for ant baits to deliver data on production and use and monitoring data on emissions at the points of use. It in concluded that in phasing out a substance, it is necessary to understand the functional characteristics of the substance in that specific application and to follow a case by case approach. This enables to find alternatives and to define the proper measures. A multidisciplinary approach is indispensable in this phase-out process.

261 Experiences of "Substitution in Practice"

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Within the research project SUPFES (Substitution of per Fluorinated compounds to Eliminate diffuse Sources), research on the substitution of hazardous chemicals in consumer products is focused on identifying feasible solutions with better sustainability performance. Such substitution model, suggesting evaluation for both technical as well as environmental and health performance, requires an interdisciplinary approach to create and/or identify feasible alternative solutions. The SUPFES SIP model include 1) characterisation of PFAS in use and for reference selected consumer products 2) initial alternative selection based on matching function criteria and toxicity and exposure assessment and 3) final selection of alternatives undergoing full environmental impact and technical performance assessment for specific scenarios. The SUPFES project has demonstrated an iterative model for practical substitution where in addition to evaluating the environmental and health performance of alternatives, the technical and economical performance are also included. It is clear that distinct substitution strategies will be required for PFASs in different textile products because of the range of performance requirements. Evaluating functionality of the different products revealed that it is critical to have a chemical (alternative) product that has the required functionality, but also to measure function in new ways matching the actual requirements. Hazard of a substitute can be reduced by improving the limits of exposure (e.g. the threshold for each chemical might be determined based on the in vivo or in vitro test and the actual exposure being lower than this threshold). This also shows that toxic compounds may be present in an alternative chemical formulation, but the levels are either really low or absent in high quality products. Furthermore, there is clear lack of key information on what is in chemical products and what is released from these products (e.g. do we have polymer degradation leading to toxic degradation products or not). From the environmental and health assessments, the specific effect of exposure to substitutes and the evaluation and guidance for consumers are needed. A need to make a trade of between protecting the user of the garment in certain working environments and high environmental impact. In addition, different environmental impact categories might give contradictory decision support.

262 Implementing a life cycle perspective in chemical alternatives assessment - the case of per- an polyfluoroalkyl substances in textile applications

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Informed chemical substitution is about eliminating chemicals that give rise to unacceptable (eco)toxicological risks, while avoiding problem shifting within a product’s or chemical’s life cycle, or between types of impacts. For this reason, the life cycle perspective becomes crucial. Chemical alternatives assessment (CAA) performance is increasingly in the focus of practitioners and life cycle assessment (LCA) and life cycle thinking are part of the more comprehensive CAA methods available. However, more detailed guidance is lacking and few practical examples have been published. A substitution case of current relevance is the phase-out of hazardous per- and polyfluoroalkyl substances (PFAS) from durable water repellent (DWR) textile applications. Alternatives are sought which offer sustained technical performance but an improved environmental and human health profile compared to the hazardous PFAS. To support an informed substitution of hazardous PFAS, and complement our previous hazard assessment, we have conducted an LCA to compare environmental and human health impacts across DWR alternatives on a
An existing shotgun to enable the use of steel gunshot. However, major gun manufacturers have confirmed that the vast majority of modern shotguns can fire alternative shot materials without any problem. In rare cases, a very old shotgun may need to be replaced or the hunter needs to use the more expensive bismuth or tungsten shot. The analysis of alternatives indicates that the use of alternatives to lead gunshot for hunting and shooting in wetlands is technically and economically feasible. While the availability of such alternative gunshot may currently vary across the EU Member States, it can be expected that a rise in demand triggered by an EU-wide regulatory action will be met on the supply side.

Delivering innovative products and solutions to the market is a driver for research and development. Drivers of innovation include changes in the market demand or the availability of new technology. New substances or new products are continuously being evaluated for their performance and functional and safety in a given application. Adaptation to the market demand thus often leads to substitution in the use of one substance for another providing improved functionality. Beyond the evaluation of the product’s safety throughout its life-cycle, increased regulatory pressure such as possible changes in hazard classification plays a role in the selection of alternative candidate substances. In a limited number of cases, the evaluation of alternatives is required by regulatory frameworks, such as the authorisation process under the REACH regulation. However, substances of very high concern (SVHC) are identified solely based on their hazard profile (e.g. CMR or PBT properties), and do not consider findings from the overall safety assessment which includes an assessment of risk. Substitution driven by the hazard characterisation alone raises a number of questions in the search for alternative substances. Relevant candidates need to demonstrate equivalent or enhanced performance and functionality in the same range of applications and demonstrate a superior environmental and health profile based on their hazard properties. However, there is a concern that substitutions based on hazard criteria may lead to regrettable substitutions, for example when it is unclear if a substitution presents a benefit in terms of overall risk to human health and the environment. Comparative risk assessment may prove to be complex as the substances typically do not have the same level of testing information to characterise the hazard. In addition, exposure may occur at greater levels when the uses require higher use rates or if processing and handling lead to higher exposures. Finally, identifying a candidate substance providing a broad range of functionalities allowing substitution in all ammunition applications in one study to meet value chain requirements in terms of performance and EH&S profile. Big data analysis in ecotoxicology: how to get new information out of existing data?

EDAPHOBASE - soil biodiversity data warehouse and its applications in ecotoxicology

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In this talk we present the soil-zoological information System EDAPHOBASE, a taxonomic-ecological database system, developed within a joined research project funded by the German Federal Ministry of Education and Research (BMBF). It combines existing taxonomical primary data on soil organisms from collections, scientific literature and reports in a data warehouse. Up to now EDAPHOBASE contains more than 500000 observations, about 300000 sites, an 140000 taxa. Data can easily be imported, quality checked, published, queried and analyzed via a web application interface. Detailed analyses can be performed with the interactive web application EDAPHOSTAT which allows species-level analysis as well as definition of reference communities. Future development of EDAPHOBASE towards a pan-European EDAPHOBASE network will focus on (1) the development of a harmonized tool for the evaluation of ecological soil quality, (2) the collection and usage of existing data from different parties all over
Europe, (3) the provision of a reference base of the ecological quality of soils and (4) the coverage of relevant needs of as many as possible European policies. Finally, necessities for practical use in common agricultural policy, circular economy and for EU transboundary issues are discussed.

267 Divining into REACH database with Rstudio to produce input data for the USEtox model for thousands of chemicals
In the context of the EU commission product environmental footprint activities (PEF) [1,2], the potential impact of chemicals emitted during the life cycle of a product is assessed via the USEtox multimedia fate model [3]. For each single chemical, this model requires dozens of physico-chemical parameters as well as data on ecotoxicity to freshwater aquatic life and toxicity to human for cancer and non-cancer endpoints. For PEF these data are required for thousands of chemicals using the most up-to-date information [4,5]. The EU commission Joint Research Centre has obtained from the European Chemicals Agency (ECHA) data on more than eight thousand chemicals. These data includes all the physico-chemical properties (166'926 test results), ecotoxicity (305068 test results) and human toxicity data (41'381 test results) available in the IUCLID 5.5 database (as of May 2020). The data was divided into eight chemical classes and eight terrestrial classes, using unique chemical properties and toxicity indicators for thousands of chemicals. The present paper focuses on the use of REACH data to calculate chemical Effect Factors. All the REACH registration data on physico-chemical, aquatic ecotoxicity and human toxicity were exported from the IUCLID 5.5 database into individual Excel files. Each Excel file was imported into the R-studio program [6] where data treatments / manipulations / calculations were performed. This allowed us to build in step wise manner until we obtained the desired selection without impacting the structure of the original file (Excel). The final code can be released to the scientific community to be reapplied on the original files obtained from the REACH database. The ecotoxicity data extracted from the IUCLID database contained about 7500 substances covering both mono and multi-taxa as well as UVCB (Unidentified Variable Composition, Complex reaction products or Biological materials). The database covers acute and chronic toxicity tests for various organisms with about 30'068 End-point study reports (ESR).

268 The effect of modelling decisions on macroinvertebrate sensitivity modelling
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Main challenges in modern ecological risk assessment (ERA) lie in the simultaneous occurrence of species diversity and compound multiplicity. The recent development of trait-based sensitivity models has proven to be successful in tackling this problem. However, this methodology is one of the first of its kind, and has to be scrutinized in detail, paying especially close attention to the many potential facets of this method in a critical manner. We start by implementing the original method, which will be used as the null method when comparing the different modelling alternatives. As input data, a toxicity, chemical classification, chemical characteristics, and a traits database are used. First, the relative sensitivity of each species to each compound is calculated. Next this relative sensitivity is averaged over all compounds belonging to the same Mode Of Action (MOA) class, resulting in a Mode Specific Sensitivity (MSS) value. Subsequently, exhaustive multiple linear regressions are made between MSS values and species traits, looking for traits which are best in explaining species sensitivity. The next step is to see the effect of different modelling decisions. As a first aspect, the effect of the used model selection criterion is studied. This is done by comparing the cross-validation error resulting from using the adjusted R² or the Akaike Information Criterion (AIC). As a second aspect, the effect of the used taxonomic level during species-trait matching is studied by comparing the adjusted R² of models resulting from species-trait matching done at family or at genus level. Third, two methods for exploiting species traits data are explored, trying to find out whether using the initial values was a treatment R allowed us to build in step wise manner until we obtained the desired selection without impacting the structure of the original file (Excel). The final code can be released to the scientific community to be reapplied on the original files obtained from the REACH database. The ecotoxicity data extracted from the IUCLID database contained about 7500 substances covering both mono and multi-taxa as well as UVCB (Unidentified Variable Composition, Complex reaction products or Biological materials). The database covers acute and chronic toxicity tests for various organisms with about 30'068 End-point study reports (ESR).

269 New approach facing new challenges in Ecotoxicology: D counter
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Routine tests in Ecotoxicology are simple, relatively inexpensive and rapid methods. They can be used to compare the sensitivity of various bioassays to chemical pollutants but data are globally missing for marine organisms. In addition, studies have mainly been focused on only one species under the same exposure, but toxicity exposure involving several species are scarce. D counter is an innovative device that can be used in ecotoxicology assays involving not only one, but also two or more different species, and proving separated data from each of the species coexisting under simultaneous exposure, whenever chronic differentiation can be achieved among the species. D counter has been mainly pointed to organisms with body sizes from 0.2mm to 3mm, but it also has been tested with larger organisms, and it can be applied either to freshwater, estuarine or marine species, being suitable in bioassays using for example, the brine shrimp Artemia salina nauplii (within $48 \text{h}$ of hatching) or neonates (less than 24 h old) from Daphnia magna or D. longispina, or even Acartia tonsa nauplii. How it works? - First it is necessary to extract the characteristic signal from sets of organisms belonging to each of the species involved in the study; this is done with a software based pattern identification and recognition procedure using training sets of organisms of each specific species, which will provide the tools for pattern recognition in the subsequent tests. The dedicated software will proceed with pattern recognition and differentiation, providing counting and body length for every single organism under test, either being from different species, presenting sub-totals - by species - and/or total counting when required. The application of this device in bioassays does not alter the requirements of the standardized methods. The device is easier to apply, saves time and it adds accuracy and objectiveness, comparing the sensitivity of various bioassays involving a single or several species, being tested independently or in mixed sets of organisms from more than one species.

270 Ceriodaphnia is eusensibile to Daphnia and should fulfill invertebrate regulatory toxicity requirements
K.A. Commong, S.E. Belanger, The Procter & Gamble Company / Environmental Stewardship and Sustainability Organization
The OECD 202 Acute Daphnia or Ceriodaphnia Toxicity Test requires the use of Daphnia magna or another “suitable Daphnia species... (e.g. Daphnia pulzii)”. The zooplankton Ceriodaphnia dubia is not considered a standard test species for chemical registration in Europe despite the availability of ISO and USEPA standard acute and chronic test methods and its wide use and acceptance in other countries. Standard acute assays conducted with C. dubia submitted to fulfill REACH dossiers can only be used as suppressing or weight of evidence studies and not as key studies. Then, simply present the battery of exposures (mixed species or single) to the device by just pouring the (tens of) flasks to the serialization component in the D counter device. The dedicated software will proceed with pattern recognition and differentiation, providing counting and body length for every single organism under test, either being from different species, presenting sub-totals – by species – and/or total counting when required. The application of this device in bioassays does not alter the requirements of the standardized methods. The device is easier to apply, saves time and it adds accuracy and objectiveness, comparing the sensitivity of various bioassays involving a single or several species, being tested independently or in mixed sets of organisms from more than one species.

271 Poster spotlight: TU001, TU002, TU003
Environmental effects of metals: Improvements to risk assessment by considering speciation and bioavailability (II)

58 SETAC Europe 28th Annual Meeting Abstract Book
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Assessment and management of stormwater on sediment recontamination due to metal contaminants
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There is a lack of understanding on the association of diffuse sources, such as episodic storm events, in the metal recontamination of sediments. The study objective is to define the effect of metals associated with storm events, with regards to the sediment recontamination in the Paleta Creek near Naval Base San Diego (NBSD), in California. Two storm-events were captured for particle size characterization and metal analysis. Receiving and outfall water were monitored for sediment deposition, contaminant water, and porewater, using auto-samplers which were triggered at each location during two different seasons. Sediments collected in outfalls, deposition traps and sediment deposits were also subjected to chemical analysis. The fractionated water and sediment samples were processed for metal extraction using the modified EPA method 3050A and 3050B, respectively, and were analyzed using ICP-MS and MERX-T. The samples were analyzed for a variety of metals, including Cd, Pb, Cu, and PbO, as a function of particle size and metal species. The percentages of clay, fine silt, coarse silt and sand, which represent particles that could be most directly related to recontamination potential. The results show that over the time the contaminant loadings decreased due to reduction in particulate contaminants while the concentrations in fine and dissolved fractions remained relatively constant and were associated with the largest particle sizes in stormwater, but only Cd is strongly associated with sediment recontamination. Cu, Pb, and Ni are associated with the dissolved phase, fine silt and clay in stormwater and present moderate impact on sediment recontamination. In addition to showing a greater dissolved fraction it appears that the depositing loads are more influenced by resuspension and redistribution of sediment than stormwater. The THg load is relatively small, stormwater recontamination does not add appreciably to sediment THg loads. The particle associations in stormwater along with spatial distribution in sediment traps can identify sources, contributing locations and effective remedial approaches. The implications of the study, can be development of identification tools that give information about the potential mobility-transport of the metals during storm events, identification of contributing locations, effective remedial approaches, and thus, help to propose best practices for stormwater and sediment management.

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The effect of percolation and form on lead bioavailability and toxicity to Enchytraeus crypticus
L. Zhang, VU University Amsterdam / Animal Ecology; C. van Gestel, Vrije Universiteit Amsterdam / Ecological Science

In the standard toxicity tests, metals are spiked freshly into test soils as easily soluble metal salts. This may lead to an inaccurate estimation of metal toxicity in soil, as it may not mimic the fate of metals in contaminated fields while the counterion could also have a toxic effect on soil organisms. The present study was set up to investigate the bioavailability and toxicity of lead nitrate (Pb(NO3)2) and lead oxide (PbO) to the potworm Enchytraeus crypticus freshly spiked and 18-months aged soil. Lufa 2.2 soil, with and without leaching. Survival and reproduction after 21 d exposure were related to total, 0.01 M CaCl2 extractable and porewater Pb concentrations in the soil and internal Pb concentrations in the surviving animals. For all treatments, 0.01 M CaCl2 extractable and porewater Pb concentrations showed a slight decrease after percolation (Pb(NO3)2) was more toxic to the enchytraeids than PbO, both for survival and reproduction and in both freshly spiked and aged soils. LC50 for the effect on enchytraeid survival, based on total Pb concentrations in the soils, did not differ for PbO after percolation in freshly spiked soils and aged soils, but increased from 1380 and 500 mg Pb/kg dry soil to 1521 and 608 mg Pb/kg dry soil in freshly spiked soils and aged soils, respectively for Pb(NO3)2. LC50 based on 0.01 M CaCl2 extractable Pb concentrations presented an increase from 2.07 and 1.72 to 2.78 and 2.42 mg Pb/kg dry soil after percolation in freshly spiked soils and aged soils for Pb(NO3)2, and a slight decreased from 2.79 and 2.45 to 2.16 and 2.18 mg Pb/kg dry soil after percolation in freshly contaminated soils and aged soils for PbO. LC50 values related to internal Pb concentrations (PbO), both Pb(NO3)2 and PbO, and ranged from 75.6 to 81.1 mg Pb/kg dry body wt in all treatments, indicating that survival of E. crypticus was better expained from internal Pb concentrations in the worms than from total or available Pb concentrations in the soil. In general, percolation did not affect total or Pb availability in the soil for Pb(NO3)2 suggesting that the counterion might have influenced Pb toxicity when Pb salts were used in the standard toxicity tests. Thus, leaching the contaminated soils before testing or using the oxide form of metals might be good ways to get rid of the influence of counterions and increase environmental realism of laboratory toxicity studies.

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To leach or not to leach: Soil enzymatic responses to metal mixture species
F. Awuah, University of Saskatchewan / Toxology Centre; S. Siciliano, University of Saskatchewan / Department of Soil Science; B.A. Hale, University of Guelph / School of Environmental Sciences

Presentation Type: Presentation preferred Abstract Title: To leach or not to leach: Soil enzymatic responses to metal mixture species Authors: F. K. Awuah1, B. Hale2 & S. Siciliano3, 1University of Saskatchewan, Toxology Center; 2University of Guelph, School of Environmental Sciences. Abstract: In soil laboratory experiments, metal mixture studies are usually carried out with metals dosed as salts, followed by leaching with artificial rainwater to remove excess salts. In the leaching process, metals are lost unequently, which affects the ratio of the mixtures in the soil. An efficient way of carrying out metal mixture experiments is by using the fixed ratio ray design. This design reduces the amount of experimental effort and allows the estimation of both additivity and interactions. In using this design, metal concentrations should be fixed in specific ratios, but this is compromised when soils are leached. Hence, an alternative method of dosing that allowed fixed ratio testing had to be determined. Two proposed alternatives were metal oxides and spinel minerals which were both abundantly found in aged salt spiked soils and field metal contaminated soils. The toxicity of the oxides and minerals to soil enzymes were tested and compared to the salts. The experiment was conducted with three Canadian soils (pH: 3.5-7), three metal species, five fixed metal(loid) mixtures, and five metals (Pb, Cu, Co, Ni, Zn) at one dose. The activity of the soil enzymes ammonia monoxygenases, beta-glucosidases, acid-phosphatas and arylsulphatases were determined colorimetrically. Results showed that leaching alone significantly inhibits the enzyme ammonia monoxygenases in all three soils. The response of acid phosphatases to the metal mixture rays followed known paradigms of bioaccessible concentrations defining toxicity. However, the response of ammonia monoxygenases was mainly due to leaching of Cd and Pb(OMe)3 from the three soils. Here, ray toxicity was highest in the soil with a pH value of 5 and vice versa for pH 3 and 7. Generally, metal salts were the most toxic form, and the spinel minerals were the least toxic. Metal oxides were chosen as a replacement for carrying out metal mixture studies in soils because no leaching was required and it was more toxic than the minerals. Keywords: Fixed ratio ray, metal oxides, spinel minerals

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Soil moisture influences the avoidance behaviour of Folsomia candida and Enchytraeus crypticus in metal(loid)-contaminated soils
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This study aimed to assess the effects of soil moisture content on the avoidance behaviour of the soil invertebrate species Folsomia candida (arthropod) and Enchytraeus crypticus (soft-bodied oligochaete) in metal(loid)-contaminated soils. Two metal(loid)-contaminated soils from Central Portugal were selected as test soils (mining soil with pH~5.9; agricultural soil with pH~4.8). Avoidance behaviour was evaluated in two-section vessels for 48 h at 20 °C. Lufa 2.2 soil was used as control soil. Avoidance tests were performed at different soil moisture contents (expressed as soil water holding capacity, WHC): 50% (standard condition), 75% (to simulate floods) and 25% (to simulate droughts). Different soil moisture content combinations were tested (test soil WHC vs. control soil WHC): 1) 50% vs. 50%, 2) 75% vs. 75%, 3) 25% vs. 25%, 4) 50% vs. 75%, 5) 50% vs. 25%, 6) 75% vs. 50%, and 7) 25% vs. 50%. Porewater metal(loid) concentrations were analysed by ICP-MS in soils incubated at 50%, 75% and 25% WHC for 48 h at 20 °C. Soils incubated at 75% WHC had higher porewater metal(loid) concentrations than those moistened at 50% and 25% WHC. This was more pronounced in the agricultural soil (e.g. 2-50 fold higher Mn, Ni and Pb concentrations at 75% soil WHC). F. candida did not avoid both metal(loid)-contaminated soils when tests were performed at the same moisture content in test and control soils while E. crypticus did, but only at 50% soil WHC (no avoidance). Vanishing performance of F. candida in test and control soils the behaviour of both invertebrate species was mainly controlled by soil moisture content. F. candida had a preference for soils moistened at 50% WHC, regardless the soils were contaminated or not. E. crypticus avoided both metal(loid)-contaminated soils in all the soil moisture combinations tested (~10-100% avoidance), except when the control soil was at drier conditions than the test soils. The present study showed that: 1) porewater metal(loid) availability across soils increased with increasing soil moisture content, especially in soils with higher acidity; 2) F. candida and E. crypticus differed in their capacity to avoid metal(loid)-contaminated soils; 3) F. candida preferred soils moistened at 50% WHC, regardless soils were contaminated or not; 4) E. crypticus could avoid metal(loid)-contaminated soils but its capacity was highly dependent on soil moisture conditions.

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Manganese bioavailability in legacy contaminated soils by medieval
metallurgical wastes

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In mining areas, the development of the tailings is influenced by metal resources through mining and metallurgical activities, for instance, of lead (Pb), silver (Ag), or iron (Fe). However, they can lead to a significant environmental contamination through the emission of metal-rich particles and wastes. In the region Burundy Franche-Comté (eastern France), iron mining and metallurgical activities were dominant over the Middle-Age period, especially in the ancient district of Bournazel. Preliminary analyses highlighted anomalous manganese (Mn) concentrations in soils surrounding medieval slag heaps. Therefore, this study aims at assessing the origin and fate of this Mn using combined physical, chemical and biological tools. For this purpose, we carried out three interdisciplinary and complementary approaches: i) mineralogical characterisation of slags (identification and mapping of their composition by XRD and SEM-EDS); ii) chemical extractions for the assessment of total and available Mn concentrations in soils and iii) environmental bioavailability of Mn using toxicityokinetics (28 days in Cantorea aspersus snails exposed to soils from 10 ancient sites of slag deposit (dated from the 5th to the 11th century) or fed with slag fragments incorporated in their diet. We identified olivine (fayalite) as the main Mn carrier in slags where its concentration reaches 4.5 wt % MnO. With time, slag weathering, as testified by the low concentration of fayalite in modern smelting ashes, and the high Mn release of Mn which accumulates in soils (up to more than 8000 mg kg⁻¹). Exctractible concentrations of Mn from soils (mainly bound to organic matter and under reducible forms) are elevated and may represent a potential toxic exposure to soil invertebrates, raising the question of Mn bioavailability in soils and slag fragments. The modeling of Mn accumulation kinetics in C. aspersus snails tissues allowed to show that the following factors influence Mn bioavailability in soils: i) the Mn invertebrates and ii) the snail ability to efficiently regulate this element. Nevertheless, when animals are directly exposed to slag fragments incorporated in their diet, the physiological mechanisms of Mn management are rapidly overloaded and internal concentrations increase up to 3000 mg Mn kg⁻¹. Hence, in slags, although often considered as inert materials, Mn turns out to be bioavailable to snails, particularly through their alteration in situ (field) and in vivo (digestive tract).

277 Chemical and ecotoxicological effects of the use of residues from the pulp and paper industry for the remediation of soils degraded by mining activities

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Biomass ash and biological sludge, both residues from the pulp and paper industry, in different mixture formulations, with and without the application of mixed municipal solid waste compost (MSWC), were used to improve the quality of soils affected by mining activities (Aljustrel mine, Iberian Pyrite Belt). The experiments consisted of (i) the evaluation of the smelting ashes bioavailability, as they are an important source of toxic metals in soils, and a pot experiment, with Agrostis tenus Sibth, to evaluate the possibility of establishing a plant cover in the amended soils. The effects on soil quality were assessed evaluating: (i) soil chemical properties, (ii) plant growth, (iii) immobilization of metals in the soil; (iv) the effects of the amendments on soil eluates toxicity, using organisms from different trophic levels: luminescence inhibition of Vibrio fischeri; 24-h mortality test with Thamnocephalus platyurus; 72-hours population growth of the green microalga Pseudokirchneriella subcapitata; and Daphnia magna acute immobilization test; and (v) soil dehydrogenase activity. Contrary to non-amended control pots, it was possible to establish a plant cover with A. tenus in pots where corrective treatments were applied, but with some variability between replicates. Phytotoxicity was observed in some of the pots related to the preliminary analyses, to allow the determination of the contamination in soil quality. In comparison to non-amended soils (controls) was further evidenced by the increase in the activity of dehydrogenase. The amendments were also able to correct soil acidity, and to increase extractable P and K. However, a significant increase in the organic matter, and N content, was only possible by the simultaneous application of biological sludge and MSWC, and decreased the higher application rates of the correctives, due to a dilution effect. In general, amending the soil with biomass ash and biological sludge decreased the toxicity of soil eluates towards the organisms used. The formulation with 30% of biological sludge and 10% MSWC, and despite its beneficial chemical effects, toxicological results did not reflect this improvement, since the presence of MSWC did not promote the decrease of toxicity towards the microalgae. Further research is needed with different plant species, since Agrostis tenus showed some phytotoxicological response. In Microplastics in freshwater and terrestrial systems - fate, monitoring and biological interactions (II)
University of Bayreuth represents one of the most comprehensive measuring programs in fluvial systems regarding the number of sample sites and the analytical accuracy. Excerpts of the study, focusing on particle number, distribution of size classes, particle shapes and polymer types at the water surface of rivers in western and southern Germany will be presented.

280 Exploring the relation between plastic concentration and river discharge in an urban river
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Rivers play a major role in transport of plastic debris from inland sources into the marine environment. Presently the relevance of various individual sources and emission pathways of plastic in rivers such as wastewater treatment plants, combined sewer overflows, surface runoff and littering can hardly be quantified. Therefore plastic emission from sub-catchments are determined by integral approaches. This study examines plastic particle concentration upstream (P1) and downstream (P2) of an urban subcatchment and establishes relationships between river discharge and plastic concentration (c-Q relationship). Suspended material > 500 µm was sampled at two sampling sites in the Parthe River, (Leipzig, Germany) in the winter (P1) and downstream (P2) of an industrial area during different discharge conditions during 17 campaigns each for 24 h. Plastic material was extracted and quantified in the suspended matter using particle size fractionation, density separation and particle cleanup followed by Raman spectroscopy. Plastic particle mass and number concentration were determined and it was observed that plastic concentration and load increased in the urban subcatchment. To explain the observed concentration and load increase, plastic input in both subcatchments was related to the catchment attributes population, catchment size, urban area, and river length revealing that population determines plastic emissions. The log-log c-Q plots of total plastic mass and particle number concentrations show an enrichment pattern at P2, hence increasing concentration with increasing discharge (positive slope b of the regression). At P1 no significant c-Q relationship was observed. This indicates that in the rural sub-catchment increasing discharge does not drive an increased mobilization of plastic material. The c-Q relationship was applied to estimate the yearly plastic emission based on river discharge data.

281 Microplastic pollution in upstream river catchments
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Microplastic particles (< 5 mm) are known to pollute large lakes and river systems globally. They have been associated with wastewater treatment plants and centers of population and industry. However, the extent to which they pervade upstream catchments is comparatively underrepresented in the study of freshwater microplastic pollution. Results presented here form part of a year-long project that aims to quantify the spatial and temporal variation of microplastic pollution in rural catchments and on the UK’s rivers. Laboratory analysis of microplastics was undertaken using a combination of optical and scanning electron microscopy. This study provides a baseline of dissolved organic matter, which changes the overall partitioning between the MP and the water phase. We measured equilibrium partition coefficients and release kinetics over 240 hours. Based on experimental data and the analytical solution of the model, characteristic times of mass transfer were calculated. These are proxies for the equilibration time and can be used to assess the relative importance of the two mass-transfer processes. Results show, that mass transfer for hydrophilic compounds usually is limited by intraparticle diffusion whereas for hydrophobic compounds it is externally limited. For intermediate compounds, a shift from internal to external dominance was observed. Calculated characteristic times show that under lab conditions the overall equilibration time decreased with increasing partition coefficient while under field conditions the opposite is the case. Thus, a simple first-order approximation of mass transfer would not be enough to transfer experimental results to field conditions adequately. Rather, it is necessary to consider true intraparticle diffusion. Application of our model to different particle sizes, shapes, materials and for varying particle concentrations underlines the fundamental differences between lab and field and allows the transferability between these different boundary conditions.

Air Pollution, Biomonitoring and Human Health (II)

284 Analysis of the contribution of a coal-fired power plant to PM10 concentrations in four sites in Southern Italy
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Freshwater environments are contaminated with various compounds. In such systems, hydrophobic organic contaminants are often associated with particles such as microplastics (MP) which are ubiquitously detected and have raised concern. Therefore plastic emission from sub-catchments is comparatively underrepresented in the study of freshwater catchments. A large part of the urban stormwater runoff is treated in retention ponds prior to discharge. This study looked into the occurrence, composition and concentration of MP in stormwater ponds, aiming to evaluate the retention efficiency of MP by these systems. The results will contribute to the understanding of MP emission from urban areas, and potential impacts on adjacent environmental compartments. Seven stormwater ponds in Denmark were selected as study sites. Surface water was collected using a pumping system equipped with a 10 µm mesh stainless steel filter. Sediment samples were collected using an Ekman bottom grab sampler. The filters from the water samplings were pre-oxidized with H2O2, followed by enzyme treatment. secondary oxidation was applied afterwards. Inorganic particles were separated by density separation using ZnCl2. Sediment samples were freeze dried and incubated in SDS solution. Enzymes were then added, followed by a dilution of the material and separation by density separation using ZnCl2. After extraction, particles (from 10 to 2000 µm) were concentrated in 50% ethanol solution. A sub-sample was deposited onto a Zeiss microscope and dried. The window was scanned by micro-Fourier Transformed Infrared Spectroscopy imaging applying an Agilent Cary 620/670 system with a 128x128 pixel FPA. The software MPHunter was used to interpret spectrums. MPs were detected in water phase of all ponds. The most abundant polymers were PP, PE and PS. The highest concentration in terms of particle number was 10.8 particle/L, while the other 6 ponds ranged from 0.07 to 2.45 particle/L. For MP mass the highest concentration was 1.2 µg/L while the other 6 ponds ranged from 0.06 to 0.4 µg/L. High density polymers were also detected in some of the ponds, this is likely due to the resuspension of sediment by wind and traps of MPs by organisms. Sediment samples are still under processing and will be included in final presentation. Nevertheless, the water samples have shown that stormwater pond do not detain all MPs, particularly for low-density polymers. With the high mobility, discharges from stormwater ponds will potentially affect adjacent environmental compartments.
sites are located in the area surrounding the “Federico II” coal-fired power plant. The studied area included the territory of the Province of Brindisi, close to the coal-fired power plant “Federico II”, and in the territory of the Province of Lecce at about 26 km SSE of the power plant. The Lecce site was included to assess the impact of the power plant emissions at middle distances. The measuring sites are Lendinizio (LN), Cistermonio (CN), Tortorato (TR) and Lecce (LE). The Lecce site is part of the Observation Center of the Global Atmosphere Watch (GAW-WMO) program. Daily PM$_{10}$ samples were collected at the different sites during measurement campaigns in different seasons (summer, autumn and winter) between 2013 and 2016. Specifically, three measurement campaigns were performed simultaneously at the four sites in 2016 and the dataset was enriched with previously available data collected in 2013 and 2015 at the sites in the province of Brindisi (LN, CL, TR) for a total of 457 daily samples. Collected samples were chemically analysed to determine 19 species: the carbonaceous components (EC and OC); the water soluble ions $\text{Cl}^{-}, \text{NO}_3^{-}, \text{SO}_4^{2-}, \text{Na}^{+}, \text{NH}_4^{+}, \text{K}^{+}, \text{Mg}^{2+}$, Ca$^{2+}$; the elements Al, Si, Ti, V, Mn, Fe, Ni, Cu and Zn. Measured data was used for source apportionment of PM$_{10}$ based on a receptor-oriented model approach that integrates the results obtained using the PMF (Principal Component Analysis) and Chemical Mass Balance (CMB), with those obtained using the CALPUFF dispersion model. This approach allows to estimate the primary contribution of the power plant to PM$_{10}$ and to obtain an estimation of its contribution to secondary sulphate.

285 Air pollution toxicity: is it the right time to leave the bench for the field? A case study integrated approach

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Air pollution (AP) is recognized as the most important environmental issue affecting human health. In Europe AP is responsible for 500,000 premature deaths mainly due to non-communicable diseases and disabilities. The epidemiological associations have already evidenced significant relationship between increases in risk factors for selected human diseases and air pollutants concentration. Finally IARC has classified outdoor air pollution as carcinogenic to humans (Group 1).

During the last decades several toxicological studies have investigated the mutagenic and genotoxic effect of PAHs on non-renal cells in vivo and in vitro. Moreover, the toxic effects of many EDCs are multifactorial, acting either by direct effects on cells or by modulation of endocrine systems. In fact, these chemicals can compete with receptors or enzymes involved in the regulation of several biological processes, such as cell differentiation, proliferation, growth, and apoptosis. The aim of this research is to identify the most affected biological targets due to the interaction with a single or multiple substances, by exposure of human buccal cells to different concentration of EDCs. We are investigating the genotoxic effect of PAHs at different concentrations and the genotoxicity of several phthalates at low concentrations. Moreover, we are exploring the potentiality of using buccal cells as a model for the study of the interaction between EDCs and nanoparticles.

Among the EDCs, endocrine disruptors (EDCs) are a group of chemicals which interfere with the endocrine system, potentially leading to developmental and reproductive disorders. EDCs can alter the normal functioning of the endocrine system by mimicking, blocking, or interfering with the natural hormones. They can disrupt the balance of hormones in our body, which can lead to a variety of health problems. This is particularly important in children, as their developing endocrine systems are more vulnerable to disruption. As a consequence, EDC exposure during early childhood can increase the risk of developing chronic diseases and disabilities.

In addition, the interaction between nanoparticles and EDCs is also under investigation. Nanoparticles can act as carriers, allowing the EDCs to reach more remote locations in the body, while EDCs can alter the behavior and properties of nanoparticles, affecting their therapeutic potential. The combination of the two can lead to unexpected effects and interactions, making the study of this interaction crucial for understanding the potential health risks.

In conclusion, the study of the interaction between EDCs and nanoparticles is essential for understanding the potential health risks and for developing strategies to mitigate these effects. The use of buccal cells as a model for this study is particularly valuable, as it allows for the investigation of the genotoxic and toxic effects of EDCs and nanoparticles on a cellular level, providing insights into the mechanisms underlying these effects.

287 Air pollution and health: early biological effects in children exposed to air pollutants and genotoxic effect of PM$_{2.5}$ in different Italian towns

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Children are a high-risk group in terms of the health effects of air pollution, and early exposure during childhood can increase the risk of developing chronic diseases in adulthood. The aim of MAPEC LIFE (Monitoring Air Pollution Effects on Children) was to evaluate the association between air pollution and specific biological indicators as biomarkers and to propose a model for estimating the global risk of early biological effects due to air pollutants and other factors in children. The study was performed on 6-8-year-old children (n=1,149) living in 5 Italian towns in different seasons. Micronucleus (MN) frequency and DNA damage were investigated in buccal cells of children. Socio-demographic and lifestyle features were collected using a questionnaire. Child exposure to air pollutants was assessed analyzing PM$_{2.5}$ (chemical composition and genotoxicity) and collecting data on air quality. In winter, the 52.7% of children showed at least one MN in cells (0.44 MN/1000 cells). A significant difference was observed among the towns. In spring, MN frequency was lower in winter (0.22 MN/1000 cells) and the difference between towns disappeared. In summer, the frequency of micronuclei was classified as high (2.1%). In children and i) to propose a model for estimating the global risk of early biological effects due to air pollutants and other factors in children. The study was performed on 6-8-year-old children (n=1,149) living in 5 Italian towns in different seasons. Micronucleus (MN) frequency and DNA damage were investigated in buccal cells of children. Socio-demographic and lifestyle features were collected using a questionnaire. Child exposure to air pollutants was assessed analyzing PM$_{2.5}$ (chemical composition and genotoxicity) and collecting data on air quality. In winter, the 52.7% of children showed at least one MN in cells (0.44 MN/1000 cells). A significant difference was observed among the towns. In spring, MN frequency was lower in winter (0.22 MN/1000 cells) and the difference between towns disappeared. In summer, the frequency of micronuclei was classified as high (2.1%). In children and ii) to propose a model for estimating the global risk of early biological effects due to air pollutants and other factors in children. The study was performed on 6-8-year-old children (n=1,149) living in 5 Italian towns in different seasons. Micronucleus (MN) frequency and DNA damage were investigated in buccal cells of children. Socio-demographic and lifestyle features were collected using a questionnaire. Child exposure to air pollutants was assessed analyzing PM$_{2.5}$ (chemical composition and genotoxicity) and collecting data on air quality. In winter, the 52.7% of children showed at least one MN in cells (0.44 MN/1000 cells). A significant difference was observed among the towns. In spring, MN frequency was lower in winter (0.22 MN/1000 cells) and the difference between towns disappeared. In summer, the frequency of micronuclei was classified as high (2.1%). In children and
implementing policies of public health protection.

288 Source apportionment of PM near steel plant by electron microscopy A. Genzo. University of Salento / Dep. of Biological and environmental Sciences and Technology; M. Siciliano, University of Salento; C. Malitesta, T. Siciliano, Università del Salento

Source apportionment based on bulk chemical analysis often uses advanced statistical tools for a detailed source categorization. In contrast, in this study the source apportionment is based on properties of individual particles determined by scanning electron microscopy with energy-dispersed spectrometry (SEM-EDS). The receptor site is located near a steel plant in the Apulia Region, South Italy. A total of 5000 particles were acquired by SEM-EDS and based on the morphology and chemical composition they have been classified into the following main groups: Aluminosilicate particles; Silicium rich particles; Ca-rich particles; Biological particles; Carbonaceous particles; Soot; Kish flakes; Salts of Sodium Chloride (sea salt); Calcium sulfate; Metal particles; Secondary particles; Fe reach particles (Fe mixture and Fe oxides). All particles, which could not be classified into one of these groups, were listed as other particles. The particle groups observed in the present investigation can be assigned to different emission sources. Besides the chemical composition, information on the morphology and mixing state of the particles is helpful for discrimination of industrial emissions originating from high temperature processes from a natural soil component, for both iron-rich particles as well as silicates. In the present study, the following source categories have been distinguished: soil, industrial, secondary, biological, soot, Ca-rich particles. In industrial, soil and secondary, particles of different groups are merged. The source apportionment analysis performed with the single particle analysis let to investigate the dimensional and mass distribution of the sources in PM10-2.5, PM2.5-1, PM4 fractions, showing that the antrropic sources are mainly present in the fine and ultrafine particles, while the natural sources are characterized by coarse dimension.

289 Oxidized transformation products of polycyclic aromatic hydrocarbons in secondary organic aerosol particles A. Kramer, Oregon State University / Environmental and Molecular Toxicology; S.L. Messey Simonich, Oregon State University / Department of Environmental and Molecular Toxicology; A. Zelenyuk, Pacific Northwest National Laboratory; K. Suski, Pacific Northwest Laboratories; D. Bell, Pacific Northwest National Laboratory

Long-range atmospheric transport of polycyclic aromatic hydrocarbons (PAHs) in fine and coarse particle matter remains a global health concern as transport models predict increased levels of PAHs within the atmosphere. The oxidation of four PAHs were studied in laboratory generated α-pinene SOA experiments, Ca sulfates, in industrial, soil and secondary, particles of different groups are merged. The source apportionment analysis performed with the single particle analysis let to investigate the dimensional and mass distribution of the sources in PM10-2.5, PM2.5-1, PM4 fractions, showing that the antrropic sources are mainly present in the fine and ultrafine particles, while the natural sources are characterized by coarse dimension.

290 The role of the p38α MAPK-mediated autophagy in cadmium-exposed monogonont rotifer Brachionus koreanus H. Kang, C. Jeong, J. Lee, Sungkyunkwan University

Tigriopus japonicus (TCS) is an antimicrobial agent that has been widely dispersed and detected in the marine environment. However, the effects of TCS in marine invertebrates are poorly understood. In this study, the effects of TCS on life cycle parameter (e.g. mortality and fecundity) along with cellular reactive oxygen species (ROS) levels, GSH content, antioxidant enzymatic activities, and mRNA expression levels of oxidative stress-induced defense genes, were analyzed using model marine copepod Tigriopus japonicus. The no observed effect concentration (NOEC) and median lethal concentration (LC50) of TCS in the adult stage were determined to be 300 μg/L and 437.476 μg/L, respectively, while in the nauplii stages the corresponding values were 20μg/L and 51.76μg/L, respectively. Fecundity was significantly reduced (P < 0.05) in response to TCS at 100 μg/L. Concentration and time-dependent analysis of ROS, GSH content (%), and antioxidant enzymatic activities (SOD, CAT, GPx) in T. japonicus were significantly increased (P < 0.05) in response to TCS exposure. Furthermore, mRNA expression of detoxification (e.g., CYPs) and antioxidant (e.g., glutathione S-transferase-sigma isoforms, Cu/Zn superoxide dismutase, catalase) genes was modulated in response to TCS exposure at different concentrations over a 24 h period. Our results revealed that TCS can reduce fecundity and induce oxidative stress with transcriptional regulation of oxidative stress-induced defense genes along with the activation of the antioxidant system in the copepod T. japonicus. Based on our investigation TCS affects survival through oxidative stress with antioxidant and detoxification defense system in T. japonicus. In addition, two CYP genes (CYP3026A3 and CYP307A1) are likely to have a potential role as biomarkers in response to TCS in T. japonicus. This study will be helpful for a better understanding of how TCS affects on antioxidant defense and detoxification mechanisms in copepod.


In aquatic organisms, cellular membranes act as the final physical barrier to xenobiotics, since the membranes are in constant contact with the ambient water column that contains various anthropogenic pollutants. In this respect, the efflux activities of membrane transporters can be considered as the first line of defense to xenobiotic exposure in aquatic organisms. GTP, GSH, and cholesterol transporters, P-glycoprotein (P-gp) and multidrug resistance-associated protein (MRP) are ATP-binding cassette (ABC) transporters that confer multixenobiotic resistance (MXR) via their efflux activity, which enables a variety of xenobiotics to be expelled from cells. MXR has been proposed as the first line of defense against xenobiotics. In this study, the protective roles of P-gp and MRP in the rotifer Brachionus koreanus were examined in response to four biocides (alachlor, chlorpyrifos, endosulfan, and endosulfan-s). These biocides are sensitive to the biocides, with reduced efflux activities of P-gp and MRP in the rotifer Br. koreanus were increased by biocide exposure, since the fluorescence intensities of the accumulated P-gp and MRP fluorescent substrates were lower in response to different biocides. Thus, exposure of rotifers to the four biocides resulted in increased P-gp and MRP activity. Moreover, the rotifers became more sensitive to the biocides, with reduced survival and slower population growth rates, when P-gp or MRP was inhibited. These findings suggest that P-gp and MRP are involved in the defense system to biocide exposure. Furthermore, the transcriptional levels of the genes encoding P-gp and MRP were examined to uncover the mechanism by which MXR is induced. Taken together, our results demonstrate that the MXR efflux system in the defense response to biocides, thereby providing a better understanding of rotifer defense mechanisms on the molecular level.

292 CRISPR/Cas9 genome editing generates Daphnia magna (loss of function) mutants for TRH and ABCB1 genes. Implications for (eco)toxicological testing. C. Rivetti, IDAEA CSIC Barcelona / Environmental Chemistry; B. Campos, Unilever R&D / Environmental Chemistry; B. Pina, IDAEA-CSIC / Department of Environmental Chemistry; H. Watanabe, Osaka University / Biotechnology; Y. Kato, Osaka University / Department of Biotechnology; C. Barata, CSIC / Environmental Chemistry

Unravel the toxicological mode of action of pollutants to non-target keyestone species may allow us to model and predict environmental risks of similar acting chemicals. OMICS technologies approaches developed in model
ecotoxicological species have allowed us to determine the mechanisms of action of many chemical contaminants. There is, however, the need for validated physiological studies applying reverse genomic tools. Here we present results on six CRISPR-Cas9 mutated Daphnia magna clones: three of them bearing mutations on the tryptophan hydroxylase gene (TRH), the rate limiting enzyme of serotonin synthesis, and another three having the transporter protein gene ABC11 mutated. Bi-allelic del TRH mutants lack serotonin and have their growth rates impaired. Bi-allelic indel ABC11 mutants had lower transcription activity. Chronic toxicity tests with the selective serotonin reuptake inhibitor fluoxetine indicated that effects of this drug enhancing offspring production was independent of serotonin. Acute toxicity test indicated that the transporter ABC11 is involved in the detoxification of ivermectin and its total or partial knockout dramatically increased its toxicity. These results provide the first evidence over for the use of reverse genetics in Daphnia to study the mechanisms of action of toxicants opening new avenues of research in a stress physiology perspective. This work was supported by the Spanish Government grant (CTM2014-51985-R).

294 Assessment of Effects and Recovery of Chaoborus Populations in a Novel-Concept Microcosm Experiment

C. Gamblin, R. Cockroft, AgroChemex Environmental Ltd; W.R. Jenkins, W R Jenkins / Regulatory Affairs Ecotoxicology; S. Norman, RidgewayEco

Chaoborus larvae of Chaoborus sp. (phantom midges) which inhabit water bodies in the agricultural landscape are very sensitive to synthetic pyrethroid insecticides and are known to offer populations the most sensitive taxon in microcosm studies. Chaoborus are holometabolous dipterans and from egg hatch, larvae develop through four aquatic instars before pupation and adult emergence. A concurrent study conducted on the same site (unpublished) elucidated that the species used in the study were multivoltine so Chaoborus have an almost all-year-round potential for re-colonisation. A new type of microcosm study was conducted to assess the extent and rate of recovery of Chaoborus populations in microcosms treated with a synthetic pyrethroid. Novel elements included spatial separation of treated and control systems by a distance of 100 m and non-invasive monitoring of larvae and pupae. The test material was applied at a single rate on two occasions with a 14-day interval to ten microcosms containing predominantly egg rafts and first-overwintering fourth instar larvae of Chaoborus obscursis. Ten untreated microcosms with similar populations of Chaoborus were established upwind of the treated units and these, together with indigenous Chaoborus, served as a potential source of adults insects for re-colonisation of the treated units. Three microcosms in each group were covered with insect-proof netting to prevent natural re-colonisation in order to assess the extent of population recovery from within microcosms. The numbers and developmental stages of larvae along with the numbers of pupae and presence and numbers of egg rafts were monitored throughout the study using non-invasive methods, from the week before the first application in May 2017 to the end of August 2017. These assessment methods were successful in enumerating the larvae, pupae and extent of adult emergence over time. The findings from the study show that the early instar larvae of Chaoborus are likely to be impacted by the insecticide but that later instars were able to survive and pupate, and that adults emerge. Recolonisation was relevant to the ‘open-field’ given that the control microcosms were a substantial distance (100 m) from the treated microcosms. The results show that populations impacted by synthetic pyrethroids are re-established in less than 8 weeks after the first application.

295 Poster spotlight: TU108, TU109, TU110

Solutions for emerging pollutants - Towards a holistic chemical quality status assessment in European freshwater resources (II)

296 Linking chemical pollution and effects – How to identify drivers of toxicity?

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European water resources are contaminated with complex mixtures of ten thousands of chemicals among them many non-regulated compounds of emerging concern but also unknown chemicals. Chemical monitoring, however, typically considers only a very small fraction of chemicals focusing on 45 priority substances according to Water Framework Directive (WFD) together with some additional River Basin Specific Pollutants. These chemicals often do not explain effects in toxicity tests and impacts on freshwater communities. Thus, we suggest a consistent tiered approach to identify drivers of toxicity in complex environmental mixtures employing mass balance and multivariate statistical approaches as well as effect-directed analysis. The approach is demonstrated using several case studies in the Rivers Danube, Rhine, Meuse and Holtemme as exemplars. A specific focus is given on endocrine disruption and mutagenicity. While natural and synthetic steroids were confirmed to play a key role for endocrine disruption, the fluorescent dye Coumarin 47 has been identified as a so far unknown environmental pollutant with great anti-androgenic potency in vitro and in vivo. In a water body with direct impact of industrial effluents individual aromatic amines probably from dye production could be identified as the drivers of mutagenicity. In contrast, mutagenic effects detectable in the River Rhine receiving multiple contaminations from many sources were driven by mixture effects of industrial and natural compounds with low individual potency but strong synergistic effects when occurring together.

297 Toxic mixtures in time-the sequence makes the poison

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It is generally agreed that ‘the dose makes the poison’ – that chemicals can be toxic or non-toxic depending on their dose. This principle assumes that once a chemical is cleared out of the organism (toxicokinetic recovery), it no longer has any effect. The impact of mixtures (Brack et al. 2016b) demonstrates, however, that mixtures, toxicodynamic recovery, which can be fast or slow. We tested four combinations of substances and found a clear difference in toxicity when the exposure order of two toxicants was reversed, while maintaining the same dose. When toxicodynamic recovery of the organism was slow relative to the interval between exposures it resulted in carry-over toxicity and so caused this sequence effect. We provide evidence for carry-over toxicity amongst chemicals acting on different targets and when exposure is several days apart. It is therefore not only the dose that makes the poison but also the exposure sequence.

298 How to deal with mixtures of pollutants in water resource management?

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Chemicals in the aquatic environment do not occur in isolation but as mixtures. Their compositions, concentrations and effects are highly dynamic with regard to their temporal and spatial occurrence. Current approaches of the EU water framework directive for assessing chemical and ecological quality do not reflect the emerging challenges. The SOLUTIONS project (Brack et al. 2015, STOTEN 503:22) deconstructed the general challenge into three questions (i) How to identify priority mixtures, (ii) How to identify drivers of mixture risk, (iii) How to measure Emerging chemical mixtures (Bruckner et al. 2015, ENV 535:2018). Here we present an advanced tiered mixture risk assessment for these groups of pollutants, considering only a very small fraction of chemicals focusing on 45 priority substances according to Water Framework Directive (WFD) together with some additional River Basin Specific Pollutants. These chemicals often do not explain effects in toxicity tests and impacts on freshwater communities. Thus, we suggest a consistent tiered approach to identify drivers of toxicity in complex environmental mixtures employing mass balance and multivariate statistical approaches as well as effect-directed analysis. The approach is demonstrated using several case studies in the Rivers Danube, Rhine, Meuse and Holtemme as exemplars. A specific focus is given on endocrine disruption and mutagenicity. While natural and synthetic steroids were confirmed to play a key role for endocrine disruption, the fluorescent dye Coumarin 47 has been identified as a so far unknown environmental pollutant with great anti-androgenic potency in vitro and in vivo. In a water body with direct impact of industrial effluents individual aromatic amines probably from dye production could be identified as the drivers of mutagenicity. In contrast, mutagenic effects detectable in the River Rhine receiving multiple contaminations from many sources were driven by mixture effects of industrial and natural compounds with low individual potency but strong synergistic effects when occurring together.
PBDE in fish. We then make an attempt to expand the assessment by integration across these pollutant groups. While the definition of Quality Standards for specific pollutant groups is a step in the right direction, our analysis shows that more efforts are needed to protect humans from possible combination effects across pollutant groups. Such efforts are currently hampered by data gaps concerning common toxicities likely to arise in humans.

300 An Advanced Methodological Framework for the Identification of Priority Pollutants and Priority Mixtures of Pollutants in European Freshwaters

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We present a proposal for an advanced methodological framework for identifying priority pollutants and priority mixtures of pollutants in European freshwaters. The proposal was developed by the EU project SOLUTIONS. It aims to tackle major shortcomings of current prioritisation procedures under the EU Water Framework Directive (WFD). While high data availability for some components may be reduced to few mixture components or even one single component which can be demonstrated to explain most of the overall risk, so-called drivers of mixture risks. Wherever conclusive evidence on significant risks and needs for risk reduction cannot be reached because all possible LOEs are somewhat blocked by significant data or knowledge gaps, mixture components of potential concern are not left unnoticed but they are prioritised for further research and testing. Some elements of the advanced methodological framework may be readily applicable under the existing WFD. Full implementation, however, requires changes in the legal text, as detailed in Brack et al. 2017 (Sci Total Environ 576:720-737).

301 A diagnostic toolbox for ecological effects of pollutant mixtures: a case study application using in situ experiments with microbial communities

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A toolbox for the detection of ecological impacts of chemicals was developed using a statistically supported, transparent and formalized weight of evidence (WOE) approach. It integrates four lines of evidence (LOE’s): (i) predictive mixture modelling, (ii) effect-directed analysis (EDA), (iii) in situ tests, and (iv) field-based monitoring studies. A systematic and quantitative method was developed for the aggregation of multiple in situ tests into LOE III, using an aggregated response index, which we termed the “average biomarker response” (ABR). The results of the four separate LOE’s are finally integrated using a systematic decision matrix that provides the main overarching conclusions that can be drawn from a given set of data and that pinpoints to critical data gaps. Here we first present an overview of the toolbox. Afterwards, we present a case study that used in situ experiments with phototoxicant biofilms (periphyton) in wastewater impacted streams. Chemical-analytical profiles initially showed clear differences of the micropollutant load in the water up- and downstream of the entry point of a sewage treatment plant effluent. These chemical-analytical data were evaluated for their potential ecotoxicological effects using a combination of toxicological approaches. Based on outcomes of this LOE, we hypothesized that clear ecological effects on the structure and function of the exposed microbial communities should be present. Indeed, these were then confirmed using the concept of pollution-induced community tolerance (PICT). In the end, the study allowed us to demonstrate that (i) the STP effluent actually caused ecological impacts on the exposed microbial community; (ii) a subsequent upgrade of the STP plant with active carbon filtration led to a recovery of the community that was driven by a lowered overall toxic pressure, (iii) PSII inhibitors were the mixture toxicity drivers, and (iv) that ecologically relevant effects go beyond a mere toxic unit summation. The presented work was a joint effort of the EU funded project SOLUTIONS, the ERAfresh project that was funded by the Swiss Federal Office for the Environment, and the IMPROVE project, which is funded by the Swedish Research Council.

Derivation, Validation and Implementation of Environmental Quality Benchmarks

302 Questioning annual average concentrations for plant protection products - TKTD modelling of real exposure profiles

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The EU Water Framework Directive defines two environmental quality standards for assessing the chemical status of surface waters. The MAC-EQS defines the Maximal Allowable Concentration that should never be exceeded and the AA-EQS defines a concentration that should’t be exceeded by the Annual Average concentration. While the MAC-EQS should account for the acute toxicity of a substance, the AA-EQS is based on chronic-ecotoxicity studies. For substances with highly fluctuating environmental concentrations like plant protection products the use of the annual average is disputed. Hence, in Switzerland it was suggested to use 14-day time-weighted average (TWA) concentrations for assessing compliance with quality standards for chronic toxicity. This approach is based on the average duration of chronic ecotoxicity tests and Haber’s rule. We assess the suitability of this approach for retrospective risk assessment by applying toxicokinetic-toxicodynamic (TKTD) modelling on high resolution exposure profiles of plant protection products measured in Swiss streams. The TKTD modelling is a proxy for the actual time-course of toxicity under time-variable exposure and is based on 7 species, 7 substances and 5 exposure profiles from 5 streams. The results confirm the suitability of the time integral of 14 days. The prediction of actual toxicity for the most toxic periods is very consistent with the toxicity modeled for the TWA. The deviations are on average less than factor 2 for each organism group tested. In addition to mortality for crustaceans and fish, only a small selection of sub-lethal effects was considered, namely reproduction and growth in water fleas and growth in duckweed and algae. The results also show that comparing quality criteria for protection against acute effects (MAC-EQS) to time-proportional 3-day mixed samples is appropriate.

303 Revision of 62 Environmental Quality Standards - lessons learned

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Environmental Quality Standards (EQS) are ecotoxicologically based thresholds that aim to prevent chronic effects of pollutants on ecosystems. Similar values exist in Switzerland and the Federal Office for the Environment recently commissioned the revision of existing EQSs for 62 substances to ensure that they are based on the current state of science. This study aimed to analyse the underlying reasons for numerical changes of EQSs and to highlight knowledge gaps. As for the original EQS derivation, relevant data were retrieved from databases, the public literature, and registration dossiers, and were, in some cases, provided by manufacturers. The reliability and relevance of ecotoxicological data were assessed using the CRED method. EQS derivation then largely followed the EU Technical Guidance for Deriving EQS. After the revision, 60 AA-EQSs and 58 MAC-EQSs were proposed. The EQS revision did not generally lead to either lower or higher EQSs. AA-EQSs increased in 13 cases (max./median fold change +9.6/3) and decreased in 18 cases (68-fold) and MAC-EQSs increased in 2 cases (50/10) and decreased in 9 cases (2.72/4). Most EQSs were derived deterministically, using the assessment factor (AF) method. Due to an increase in data for some
substances, the number of AA-EQs and MAC-EQs derived using Species Sensitivity Distributions (SSDs) increased from 2 to 5 and from 7 to 10, respectively. For AA-EQs derivation, AFs were reduced in 12 cases and increased in only 6 cases. For the MAC-EQs derivation, AF were reduced in 5 cases and increased only in one case. Our study demonstrates that EQSs based on small data sets are more prone to large numerical changes when revised. Hence, an update often reduced uncertainty associated with the derived EQVs, as evident from application of lower AFs and more frequent EQS derivations based on SSDs. This is likely to make EQSs more robust against larger changes in future revisions. Nevertheless, for the majority of the substances considered in this study, data sets were insufficient to construct SSDs. This is mostly due to a lack of studies using non-standard test species and species from important taxonomic groups, such as anurans and insects. Animal types, species and the mode of action of a substance, this factor alone prevented the use of lower AFs. Finally, recommendations regarding assessability and quality of ecotoxicity data from industry studies and from the scientific literature are presented.

304 Endocrine disrupting properties: how far and consistent they are considered deriving Water Framework Directive Environmental Quality Standards? A case study tackling French and EU EQS values

A. JAMES-CASAS, INERIS / Toxicological Ecotoxicological Assessment of chemical Substances ETES; A. BOTHAMY, INERIS; S. ANDRES, INERIS / Toxicological Ecotoxicological Assessment of chemical Substances TI: A study on endocrine disrupters: Group 4 contains all the substances that should be tackled in diverse regulatory frameworks, among which the Water Framework Directive Common Implementation Strategy (WFD CIS). In this context, endocrine disruption (ED) is quoted several times as an issue for deriving water quality thresholds in the European Commission Technical Guidance for Deriving Environmental Quality Standards (TGD-EQS). However, even if this guidance indicates that ED properties as a reason for growing concern it does not properly recommend any specific methodological approach to consider these properties while deriving EQS values. In order to evaluate the usefulness of such a recommendation and the extent to which it should be implemented, a state of the art of how ED properties are currently been taken on board in the EQSs already derived at EU and national level was made. To begin with, the work consisted in carrying out an inventory of substances for which an EQS had been derived and a factsheet describing the reasoning behind value was available. These substances made up the universe of 178 substances on which further work was led. Then, an analysis was made of these substances EQSs derivation to categorise them according to how ED properties where reported and taken on board if necessary for protection of wildlife and human health. This work led to 4 groups of substances. Group 1 contains substances whose EQS values do not consider ED effects, and which need to be reassessed as a matter of priority. Substances for which EQS derivation has considered ED effects but whose rationale does not clearly explain this are grouped together in Group 2 and should be verified. Group 3 corresponds to substances whose ED characteristics have been considered by an additional safety factor and / or EFs. Group 4 contains all the other substances so no ED effects demonstrated from now on. No action is required for these last two categories. This state of play and categorisation work made it possible to prioritise substances for which EQS should be updated first as regards their ED potential. Also, this work gives more insight in how to derive EQSs as regards ED potential in order to further propose recommendations for a harmonisation of the methodology in the future.

305 Bringing water quality benchmark derivation approaches into the 21st century

R.A. van Dam, Environmental Research Institute of the Supervising Scientist / Department of the Environment and Energy; G.E. Batley, CSIRO Land and Water / Centre for Environmental Contaminants Research; R. Fisher, Australian Institute of Marine Science; D. Fox, Environmetrics Australia; A.I. Harford, Environmental Research Institute of the Supervising Scientist / Dept. of the Environment and Energy; C.L. Humphrey, Environmental Research Institute of the Supervising Scientist / Department of the Environment and Energy; A. Peters, Department of the Environment and Energy; J. Stauber, CSIRO / CSIRO Land and Water; M.S. Warne, Coventry University / Centre for Agroecology, Water and Resilience

The most common method for deriving water quality benchmarks (WQBs) for toxicants is the use of a species sensitivity distribution (SSD) to estimate a chemical concentration, with an uncertainty range of 95% of species. Although variations exist in the specifics of the methods employed by jurisdictions around the world, the fundamental SSD approach is similar and, moreover, has not changed markedly over the past 20 years, despite a significant body of published research aimed at improving or developing new derivation methods. The recent revision of the Australian and New Zealand SSD-based derivation method has re-highlighted numerous limitations of the SSD approach for certain data situations and toxicant types; for example, small sample sizes, model choice and fit, and accommodating different routes of exposure (e.g. for persistent, bioaccumulative and toxic compounds) and specific mechanisms of toxicity (i.e. bimodality). However, areas for improvement of WQB derivation methods extend beyond just refining SSD-based approaches, to the use of non-SSD approaches and weight of evidence approaches that give consideration to both laboratory- and field-effects data. Other opportunities for improvement exist in the acquisition of data for WQBs (e.g. type and acceptability of toxicity data), as well as the application of WQBs in water quality management. Thus, it is important to identify and target the limitations that, if addressed, will yield the biggest benefits to environmental protection. Experience has shown that a ‘one size fits all’ approach to WQB derivation does not work, and an alternative is to provide flexibility and adopt approaches that do the best job in the face of the specific conditions and uncertainty posed by different situations. However, this may increase the complexity of the derivation process and, thus, decrease understanding and adoption by users. This presentation will examine a variety of questions associated with WQB derivation and the motivations for and against what research efforts have been undertaken to significantly evolve the way we derive WQBs? To what extent have the outcomes of this research been adopted in formal derivation methods? Why has the fundamental SSD approach not changed markedly over the past 20 years despite various proposed improvements? Do we need to move to a new generation of WQB derivation approaches and, if so, what are the aspects that will have the biggest impact, and the criteria that would make new approaches amenable for adoption?

306 The quest for consistent environmental protection: the challenge of variable water quality guidelines between regulatory jurisdictions

G. Merrington, A. Peters, wca; S. Kosmala, WCA Environment Limited

One of the tools used by regulatory jurisdictions to deliver environmental protection is Water Quality Guidelines (WQGs) or Environmental Quality Standards. These are thresholds expressed as a chemical concentration, with an associated summary statistic (e.g. maximum) and period (e.g. “annual average”). These thresholds are used around the world for a number of purposes including to assess the water quality versus water quality criteria and through setting effluent discharge permits. Historically, most regulatory jurisdictions across the globe have, at the very least, WQGs for some trace elements. The perceived challenge for many in the regulated community, especially multinational organisations, is the lack of transparency in derivation and implementation of WQG within a regulatory jurisdiction and the inconsistent environmental protection levels between regulatory jurisdictions, despite having the same protection goals. Here we address the veracity of that perception and attempt to understand its source. There is limited consistency in environmental protection, as judged by WQG for the same trace element, across regulatory jurisdictions. The absence of robust protocols (or any protocols that can be reviewed) for the derivation is a major factor as is the incredible lack of resources allocations for many regulators to update WQG and account for new scientific developments. There are unfortunate repetitive cycles of derivation that each jurisdiction goes through for the same substances and perhaps there is benefit in sharing knowledge and understanding across jurisdictions that would deliver consistent and transparent levels of environmental protection.

307 A Call for Greater International Collaboration in Developing Environmental Quality Benchmarks: Many Hands Make Lighter Work!

M.S. Warne, Coventry University / Centre for Agroecology, Water and Resilience; K.M. Leung, The University of Hong Kong / The Swire Institute of Marine Science; G. Merrington, wca

Environmental quality benchmarks (EQBs, also variously called guidelines, standards, criteria) are an internationally accepted means of protecting ecosystems from the adverse effects of toxicants. As such, numerous countries, states/provinces, regions, academics and consultants have developed EQBs. As a result there are numerous EQBs for the same suite of chemicals (e.g. copper, lead, benzene), each slightly different. This is a huge waste of resources. These differences arise because of the differences in the methods used to derive the EQBs, which govern what is considered acute and chronic, what data can be used, and the magnitude of assessment factors etc. The current situation with the derivation of EQBs has been compared to that of toothbrushes – “everyone has one and no-one else wants to use anyone else’s” and disagreements arise about whose “toothbrush” is best, whether particular features are “necessary” and whichEQB sets have an achieved or potential influence in a region and/ or country. What is also evident is that the “toothbrushes” vary considerably, and, moreover, has not changed markedly over the past 20 years, despite a significant body of published research aimed at improving or developing new derivation methods. The most common method for deriving water quality benchmarks (WQBs) for toxicants is the use of a species sensitivity distribution (SSD) to estimate a chemical concentration, with an uncertainty range of 95% of species. Although variations exist in the specifics of the methods employed by jurisdictions around the world, the fundamental SSD approach is similar and, moreover, has not changed markedly over the past 20 years, despite a significant body of published research aimed at improving or developing new derivation methods. The recent revision of the Australian and New Zealand SSD-based derivation method has re-highlighted numerous limitations of the SSD approach for certain data situations and toxicant types; for example, small sample sizes, model choice and fit, and accommodating different routes of exposure (e.g. for persistent, bioaccumulative and toxic compounds) and specific mechanisms of toxicity (i.e. bimodality). However, areas for improvement of WQB derivation methods extend beyond just refining SSD-based approaches, to the use of non-SSD approaches and weight of evidence approaches that give consideration to both laboratory- and field-effects data. Other opportunities for improvement exist in the acquisition of data for WQBs (e.g. type and acceptability of toxicity data), as well as the application of WQBs in water quality management. Thus, it is important to identify and target the limitations that, if addressed, will yield the biggest benefits to environmental protection. Experience has shown that a ‘one size fits all’ approach to WQB derivation does not work, and an alternative is to provide flexibility and adopt approaches that do the best job in the face of the specific conditions and uncertainty posed by different situations. However, this may increase the complexity of the derivation process and, thus, decrease understanding and adoption by users. This presentation will examine a variety of questions associated with WQB derivation and the motivations for and against what research efforts have been undertaken to significantly evolve the way we derive WQBs? To what extent have the outcomes of this research been adopted in formal derivation methods? Why has the fundamental SSD approach not changed markedly over the past 20 years despite various proposed improvements? Do we need to move to a new generation of WQB derivation approaches and, if so, what are the aspects that will have the biggest impact, and the criteria that would make new approaches amenable for adoption?
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Microbial community ecotoxicology in environmental risk assessment and ecosystem monitoring

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The impact of anthropogenic activities on bacterial and viral diversity in the Eastern Mediterranean Sea
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The Eastern Mediterranean Sea is a low-nutrient low-chlorophyll marine ecosystem [1] but some variability within the basin does exist. Indeed, several coasts are influenced by anthropogenic processes, and specifically in the Greek coasts these include industrial, harbor, agriculture, mariculture activities, urbanization and tourism [2]. Our hypothesis was that prokaryotic and viral community diversity is differently affected in contrasting coastal systems by anthropogenic pressures. We used 16S rRNA gene amplicon and whole virome sequencing at stations characterized by different chemical features based on the “National monitoring project for the implementation the Water Framework Directive (2000/60/EE) in Greece” [2]. We focused on viral auxiliary metabolic genes and the influence of heavy metals (Cu, Cd, Co, Ni, Pb, Zn, Cr and Hg). Significant differences were found at the genus level between the sampling stations. Proteobacteria were dominant in all stations, while Bacteroidetes were more pronounced in some of the stations. Rare phyla were higher in Echinades and Patraikos Gulf. 16S rRNA patterns resembled abiotic variables, and especially the patterns of heavy metals Cd, Cd, Cu and Pb. The highest concentrations of NO2-, NO3-, NH4+, PO43-, SiO2 and chlorophyll a were found in stations influenced by extensive industrial, agricultural and maricultural activities. The 3 stations of Amvrakikos Gulf were highly variable in terms of community structure. Significantly lower relative abundance of Verrucomicrobia and Bacteroidetes in the “control” than in the “impact” station in Kefalonia (inside and outside the influence of the fish farms, respectively) was seen. Bacterial 16S rRNA analysis revealed significant differences between stations along the Greek coast, suggesting that each station hosts a different community. Analysis of viral metagenomes will show if community composition reflects the anthropogenic activities in these areas, and if lysogeny (i.e. viral integrase and auxiliary metabolic genes’ abundance) is a prevalent life strategy [1]. Krom MD, Emeis K-C, Van Cappellen P. 2010. Why is the Eastern Mediterranean phosphorus limited?. Prog Oceanogr 85:236-244. [2] Pavlidou A, Simboura N, Rousselaki E, Taspinak M, Papadou K, Drakopoulos P, Assimakopoulos G, Kontoyiannis H, Panayotidis P. 2015. Methods of etiopathogenesis assessment in the context of the water framework directive. Cont Shelf Res. 108: 156-168.

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Impacts of stormwater on microbial community structure and function in estuarine sediments
K. Bollington, Macquarie University / Evolution and Ecology Research Centre; P. Steinberg, University of New South Wales / Centre of Marine BiInnovation; S.L. Simpson, CSIRO Land and Water / Centre for Environmental Contaminants Research; J. Potts, P. Scanes, NSW Office of Environment and Heritage; S.C. Bärer, University of New South Wales; M. Sutherland, NSW Office of Environment and Heritage; V.X. Sim, University of New South Wales; T. Lachnit, University of Kiel; S. Swarup, National University of Singapore; S. Kjellberg, Nanyang Technological University / The Singapore Centre on Environmental Life Sciences Engineering; M. Doblin, Department of Environmental Sciences / Department of Environmental Sciences; G. Birch, Sydney University / School of Geoscience; P. Gribben, University of New South Wales; E.L. Johnston, University of New South Wales / Evolution and Ecology Research Centre. Estuaries are diverse and productive ecosystems that are subject to high levels of disturbance. They are exposed to multiple stressors such as legacy contaminants in sediments and ongoing inputs of nutrients and metals via stormwater, but we still have little understanding of the consequences for ecosystem functioning. We surveyed sediment communities at four locations with large stormwater drains in Sydney Harbour, Australia. Locations were either poorly-flushed embayments or well-flushed embayments. Sediments were collected monthly during base rainfall (<5mm/day) for 4 months from 3 sites within each location at increasing distance from the stormwater drain (0, 200 and 1000 m). We also collected sediments after 2 large rain events (>150mm in 24h). Next-generation sequencing was used to characterize the microbial community and sediment was subsampled for metals, total organic carbon, total nitrogen and phosphate. Sediment cores were also collected to assess the chemical processes including primary productivity, community respiration and nutrient cycling. We observed major shifts in the microbial community related to exposure to legacy contaminants and new stormwater contaminant inputs. We also found trends of decreasing community respiration rates away from storm drains and lowest rates of primary production during base rainfall. The results have implications for future management of stormwater in estuaries and increase our understanding of how to conserve crucial sediment community diversity and function.

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Drought as environmental driver on ciliates and micrometazoa communities in a multistressors scenario. An experimental approach
J. López-Duval, F. Romero, V. Acuña, S. Sabater, ICRAN Catalan Institute for Water Research
Climate change will affect agriculture practices and productivity because increased intensity and frequency of drought events, storms and floods will increase. Drought events and changes in phenology, that means a future increase in the use of pesticides and, in consequence higher risk of freshwater pollution. In addition climate change will lead to higher severity of drought events and higher temperatures. Ciliates and micrometazoa in freshwater ecosystems play an important role in the processing of organic matter and as basal resource for consumer organisms. The present work aims to study how these environmental and chemical stressors, and their interactions, in a future scenario of climate change can affect these communities in freshwater sediments. To study how drought (D), warming (T) and a realistic environmental mixture of pesticides (P) can affect the communities of ciliates and micrometazoa in river sediments, we developed an experiment with a factorial design in experimental indoor channels with natural sediment from a pristine river (24 channels, 3 replicates, 3 exposure conditions). The community was exposed to the stressors for 7 weeks. Diversity of Ciliophora and micrometazoa communities was studied twice during the experiment (one week before and after 7 weeks of chronic exposure). Significant changes in community composition between pre- and post- exposure were observed for all treatments. Community was dominated by micrometazoa in all treatments in terms of density, but a trend of increasing the percentage of ciliates in those treatments with stressors was observed, indicating a possible advantage of ciliates in stressed environments. At the end of the experiment total density was significantly higher respect to control in D, DP and TDP treatments (p < 0.001 Dunnet’s test) while diversity was significantly higher in D, TD and TP conditions (p < 0.001 Dunnet’s test). Taking into account that community composition for each treatment was the most important factor causing significant differences in community composition (PERMANOVA p < 0.001). The mixture of pesticides at realistic environmental concentrations did not cause any effect on the studied communities. Our results suggest that flow reduction is the main driver for changes in micrometazoa and ciliate communities in our experimental system while pesticides and temperature produce significant effects only in combination with drought.

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Linking pesticide pollution with periphyton quality in agricultural streams: a fatty acids approach
N. Corbull, University of Gothenburg / Sweden / Biological and Environmental Sciences; Håkansson G, Department of Biological and Environmental Sciences; A. Nilsson, University of Gothenburg / Section of Ophthalomyology, Dept. Clinical Neuroscience, Institute of Neuroscience and Physiology, Sahlgrenska Academy; K. Johansson, University of Tartu / Institute of Technology; H. Spångfors, Halmstad University; M. Kahler, SLU Swedish University of Agricultural Sciences / Department of Aquatic Sciences and Assessment; J. Krueger, Swedish University of Agricultural Sciences / Centre for Chemical Pesticides; T. Backhaus, University of Gothenburg / Department of Biological and Environmental Sciences
Although the environmental risks associated with pesticide pollution in agricultural streams are quite well documented, little is known about its potential effects on periphyton quality. Periphyton provides many of the essential polysaturated fatty acids (PUFA) that are needed for organisms at higher trophic levels in river food webs. This study aims to assess the effects of pesticide mixtures on periphyton quality in situ. Two streams (Höje å, Skivarpsån and M42) located in the agriculturally dominated region of Skåne (SE Sweden) were sampled in September and October 2016. The effects of pesticide pollution were assessed by passive field sampling coupled with laboratory ecotoxicity tests, by mixture toxicity modelling to predict which chemical stressors were potentially driving the toxicity, and by examining the fatty acid profiles, pigment content and algal diversity of periphyton communities. Results from water chemical analyses clearly showed higher levels of nutrients and pesticide pollution in Skivarpsån and M42 than in Höje å. Ecotoxicity tests using the passive sampler extracts demonstrated that the pesticide mixture occurring at Skivarpsån and M42 were toxic for periphyton communities from Höje å, causing an inhibition of the photosynthetic activity up to 63% and 53%, respectively. Cluster and principal component analyses based on pigments content, algal diversity and fatty acid profiles, clearly separated the periphyton from the three river sites studied. Algal biomass from periphyton of pesticide polluted streams (Skivarpsån and M42) was higher than in Höje å. The nutritive quality of the periphyton differed among streams, and fatty acids considered high-quality such as eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) were also more abundant in pesticide polluted streams (Skivarpsån and M42). Overall, even though results from the lab show that the mixture of pesticide pollution in the studied
streams might be toxic for periphyton (i.e. inhibiting the photosynthetic activity), being herbicides the driving chemical stressors. Results from the field, indicate that when the levels of pesticide pollution are low and co-occur with high levels of nutrients pollution, nutrients might mask pesticides effects on periphyton quantity and quality because compensatory effects from nutrients.

312 Estrone and triclosan mixture alters soil metagenomics during degradaton
D.L. Carv, Texas Tech University / Biological Sciences; E. Osuji, Texas Tech University / Biological Science
Wastewater derived from domestic use commonly contains mixture of pharmaceutical and personal product (PPCP), but its persistence and accumulation in the soil. The consequences for soil microbial community processes are poorly understood. Estrone and triclosan are two common PPCPs of domestic wastewater. Soil microbial communities degrade a variety of PPCPs however; most studies have only addressed single compound designs neglecting the reality of their co-occurrence in nature. In this study, we examined the interaction between estrone and triclosan mixture, their potential to persist and disrupt soil microbial community composition and function. Soil was spiked with estrone, triclosan, and a 1:1 mixture of estrone: triclosan, and incubated for 90 days in the dark at 27°C. We examined soil microbial function dynamics using commercial Biolog EcoPlates™. Microbial degradation rates were compared over the 90 days’ incubation period using high performance liquid chromatography. Metagenomic analysis by 16S rRNA was used to determine changes in microbial community over time. There was a significant increase in substrate activity and substrate richness in all treatments. Each microbial community utilized different carbon substrates by day 90 whereas they had exhibited similar substrate utilization at day 0. Estrone and triclosan as single compound treatments exhibited half-lives of 6.8 days (estrone) and 26.7 days (triclosan). The rate of degradation of the binary estrone:triclosan mixture was the same as the individual compounds. There was a decrease in species diversity between day 0 and all other treatments at day 90 with establishment of unique OTUs in each treatment group at day 90. Metagenomic analyses indicate distinct communities by treatment 90 days after exposure even though Bacillus sp. was dominant in all the day 90 treatments. Soil microbial communities are adept at degrading estrone and triclosan in this soil whether occurring singly or as a binary mixture thus preventing accumulation in soil and subsequent contamination of ground water.

313 Poster spotlight: TU014, TU015, TU016
Integrating life cycle approaches towards a sustainable circular economy (II)

314 Region-specific life cycle inventories for tailings disposal in ecoinvent v3
D.A. Turner, EMPA / Technology and Society Lab; G. Doka, Doka Life Cycle Assessments; A. Haarman, EMPA Technology & Society Lab / Technology & Society Lab; R. Hirschier, EMPA / Technology and Society Lab
Tailings, a waste material produced during mineral concentration (beneficiation), often contain significant quantities of mobile toxic metals and are typically produced in large quantities. To manage these wastes, tailings are commonly stored behind dammed impoundments, known as “tailings ponds”. These ponds pose a significant long-term pollution risk as metals may leach out into the surrounding environment, potentially over very long timeframes. The management of tailings therefore represents an important environmental burden for primary metal production. To help life cycle assessment (LCA) practitioners quantify this important environmental burden, the ecoinvent database contains – since 2009 and the release of version 2.1 – a global average life cycle inventory (LCI) dataset for sulphidic tailings disposal, developed using a dedicated tailings emissions model. However, the dataset was intended only to serve as a first generic estimate and is based on highly aggregated data that attributes an identical burden to each kilogram of waste, regardless of its composition. Given their relevance to the overall impacts of primary metals production, access to more detailed, region-specific LCI data on tailings disposal is crucial for a more comprehensive and adequate integration of primary metals in LCA studies. Here, we present an extended model for assessing the long-term emissions from tailings disposal. The model can heed a specific tailings composition and local climate data, allowing for the creation of site- or region-specific LCI data based on an extended literature survey and data on tailings compositions and leachate concentrations from different mine sites worldwide, the model was used to develop new country- and region-specific datasets for tailings disposal from a range of ore types, which will contribute to improving the quality and hence reducing uncertainties in LCA studies worldwide. Our presentation will give an overview of the extended model and related datasets, which will be integrated in a later version of ecoinvent. We will also highlight its improvements compared to the previous model by presenting the results of an LCA case study of region-specific primary metals production in order to demonstrate the important differences between global average and situation-specific calculations in such an important sector as primary metals production.

315 Closing the copper cycle in the EU-28: scenario analysis and potentials for GHG emissions reduction
L. Ciacci, Alma Mater Studiorum - University of Bologna; F. Passarini, Alma Mater Studiorum - University of Bologna /Dept. of Industrial Chemistry
Copper is widely used in modern society, finding application in traditional end-uses such as plumbing, infrastructure, and transportation, but it is also an essential material in emerging green energy technologies. Europe (i.e., EU-28) has modest natural deposits and strongly depends on imports to meet the domestic demand. In this regard, the importance and possible supply shortages, end-of-life recycling can secure access to secondary copper forms and support the implementation of a circular economy. In addition, as copper recycling is generally less energy intensive than primary copper production, closing the elemental cycle through recycling would result in significant environmental benefits. However, despite a well-established industry network in the copper value chain, the EU-28 is still far from perfect recycling highlighting wide margins for improvements. Some of these potentials for copper circularity and environmental benefits were explored combining four well-regarded UNEP scenarios with material flow analysis. For each scenario, the copper demand and supply in the region was modeled to 2050. We commented the results in the case of stationary end-of-life recycling performance and under the hypothetical implementation of a near-perfect recycling economic function as new economic indicator calculated to total cost the resulting energy savings and greenhouse gas emissions reduction. The results show that copper recycling can contribute significantly to reduce the energy requirements and mitigate greenhouse gas emissions associated with the regional copper industry. However, for three out of the four scenarios the current recycling performance seems not to be enough to close the copper cycle. Fundamental constraints are likely to limit the implementation of a circular economy unless dramatic changes occur in the current pattern of copper production, consumption and recycling at end-of-life.

316 Multi-Objective Reverse Supply Chain Network Design of Fluorescent Lamps with Piecewise Linear Functions
C. Leg, L. Pagapgeorgiou, University College London / Department of Chemical Engineering; N. Shah, Imperial College London / Department of Chemical Engineering
In this study, a reverse supply chain model has been developed to support strategic decisions making problems as associations is its second-order polynomial design and less operation. The examined networks comprise multi-echelons, including disposer markets, collection facilities, recyclings and reuse markets connected by a transportation mode. The system is modelled as a multi-objective mixed-integer linear programming (MILP) optimisation problem allowing the inherent trade-offs between the conflicting economic and environmental objectives to be explored. The economic function is not economic indicators calculated to total cost the resulting production savings. Total cost includes capital and operation costs required to operate the supply chain network. The production saving is the revenue obtained from selling secondary products. In contrast, the ecological objective function is based upon net environmental value. This is achieved by adopting the principles of LCA, expanding the network boundaries to incorporate a set of life cycle stages and using the Korean Eco-Informatics System (KEIS) for calculation of the network and avoided burdens. In addition, the environmental and economic performances of reverse supply chain networks are assessed through the development of a spatially explicit model that combines logistics and Geographic Information Systems. The model is used to address strategic decisions involving the location, number and capacity of collection and recycling facilities; selection of recycling technologies; and assignment of transportation links required to satisfy demands in terms of demand and market. At the operational level, optimal recycling profiles and flows of material between various components within the supply chain are determined. Furthermore, the model considers the economies of scale (large, medium and small) of collection & recycling facilities and transportation links, and explores whether centralised recycling is favoured over decentralised recycling. The feasibility of the proposed models is explored within a South Korea context in order to determine the optimal reverse supply chain configuration of fluorescent lamps.

317 The use of Life Cycle Assessment to adjust consumption taxes: The concept of a Damage and Value Added Tax
B. Timmermans, University Leiden /Leiden University /Geography Systems. The model is used to address strategic decisions involving the location, number and capacity of collection and recycling facilities; selection of recycling technologies; and assignment of transportation links required to satisfy returns and demand at the markets. At the operational level, optimal recycling profiles and flows of material between various components within the supply chain are determined. Furthermore, the model considers the economies of scale (large, medium and small) of collection & recycling facilities and transportation links, and explores whether centralised recycling is favoured over decentralised recycling. The feasibility of the proposed models is explored within a South Korea context in order to determine the optimal reverse supply chain configuration of fluorescent lamps.
Safe by Design: responsible and innovative research for safe and sustainable chemistry

319 Poster spotlight: TU214, TU215, TU237

320 Silica coating for the control of nano-reactivity
S. Ortelli, CNR ISTECC; M. Blosi, CNR; D. Gardini, CNR ISTECC; A. Costa, CNR Nano-titanium dioxide (TiO₂) and nano-silver (Ag) are among the materials most investigated for their technological importance and consequent interest in terms of their environment, health and safety (EHS) issues. In particular these particles cause alert for their capacity to generate, transport and release potentially toxicants such as metal ions and reactive oxygen species that can induce several negative effects, responsible for cytotoxicity. In this study we investigated silica coating as technique for control two recognised toxicity drivers for nano TiO₂ and Ag that are the exogenous production of ROS and the Ag/Ag⁺ total distribution. We evaluated the effect that silica coating had on physicochemical properties (size, shape and zeta potential), optical properties and toxicological performances. We first demonstrated that both at colloidal and dried state a matrix of SiO₂ surrounding TiO₂ and Ag nanoparicles was formed, even by simply playing with colloidal attraction between the two hetero-phases. The presence of silica coating seems to have two important effects for the control of ROS and Ag⁺ toxicants, representing a safe by molecular design solution for the control of nanotoxicity: 1) Silica acts as dispersing/gelling matrix for ROS, decreasing the production of ROS, but improving the photocatalytic performances of pristine sample; 2) Silica acts as reservoir for Ag⁺ ions, decreasing the amount of immediately available fraction and so improving the range of concentration where the sample shows antibacterial properties despite to negligible cytotoxicity.

321 Framework for the optimal design of sustainable chemical processes
A. Gonzalez Garay, R. Calvo-Serrano, G. Guillén Gosalbez, Imperial College London / Chemical Engineering
Given its natural link between fundamental science, engineering and industrial practice, the chemical industry plays a key role in meeting the challenges of sustainable development. In particular, the use of computer aided tools for the generation of sustainable processes is essential to facilitate the transition towards a more sustainable chemical industry. In this work, we present a framework that incorporates sustainability principles in the design of chemical processes. The methodology proposed uses life cycle assessment to assess the sustainability of the processes; surrogate modeling and objective-reduction techniques to enhance the optimization of the processes; and data envelopment analysis (DEA) as multi-criteria analysis tool. The use of DEA facilitates the post-optimal analysis of the Pareto front by filtering and ranking the optimal designs that conform the Pareto frontier without the need to define explicit weights. In addition, DEA provides improvement targets for suboptimal alternatives that if attained would make them optimal. Finally, the proposed framework makes the tools described above immediately available; this can be used to guide retrofit efforts towards more effective actions. The capabilities of the framework are shown in a case study based on the production of methanol from CO₂ and hydrogen.

322 A decision framework for substances of very high concern at the interface of chemicals, products and waste
Reuse and recycling of products are key elements towards a sustainable and circular economy. Besides the circular economy policy, a non-toxic environment is being pursued as well. Care should be given to the reuse and recycling of waste streams containing substances of very high concern (SVHC). Ideally, the presence of SVHCs in the design and production phase should be prevented by applying Safe-by-Design alternatives. However, we have to realize that we are still in an era in which we are faced with numerous (legacy) SVHCs in waste streams. For these waste streams, safe ‘end-of-life’ solutions have to be found in order to stimulate the circular economy and safeguard a non-toxic environment. Within this study, we develop a general framework to decide how waste streams with legacy SVHCs should be managed. The framework is specifically developed for the licensing process and provides guidance to license applicants and license authorities in the Netherlands. By following the framework, it will indicate whether the recycling of a specific waste stream into a specific end-product can be considered as acceptable with respect to the SVHC content. In principle, the use of this framework consists of three steps: 1) identification of SVHCs in the waste stream; 2) a basic decision scheme in order to decide whether a more elaborate risk analysis is necessary or whether the risks can be considered as acceptable; and 3) a risk analysis. Within the risk analysis, the acceptability of recycling will be assessed by weighing various aspects, including: availability and feasibility of SVHC removal possibilities, exceedance of SVHC concentration limit values, potential SVHC exposure of man and the environment, and the traceability of the material stream (including SVHC) during the whole life cycle. This framework is a first step to increase and warrant safe and sustainable recycling of waste streams. Future adjustments of this framework will be required in upcoming years based on practical experiences of...
licensing authorities. Furthermore, it is advised to develop a broader decision scheme that besides SVHCs also considers and weighs other risk and benefit factors of recycling, like the risk of pathogens and medicine residues and the benefits with respect to sustainability (e.g. carbon footprint). Such a development will further stimulate the transition towards a safe and sustainable economy.

323 Emissions of PFASs and alternatives from the durable water repellence layer (DWR) of textiles during use

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In the durable water repellence (DWR) layer of textiles of outdoor clothing, PFASs have been used because their perfluoroalkyl chains have the ability to repel liquids of a wide range of polarities (DWPs, hydrophobic), DWR compounds, like PFASs and silicones are emitted to air, as well as to rain water and washing water. During the use phase of outdoor clothing, DWR chemicals are emitted to the environment.

324 Chemicals in plastic packaging: Prioritization of hazardous substances

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Plastic packaging is increasingly used globally, causing raising concerns for the environment and human health from littering, release of microplastic and leakage of hazardous chemicals. Specifically, chemicals may migrate into foods or the environment during use, disposal, and recycling of the packaging material.

Occupational exposure during plastic packaging manufacture is also relevant. One of the main obstacles to assessing the risks of chemicals originating from plastic packaging is the absence of information on the materials’ exact chemical composition. In order to provide an overview of the chemicals associated with plastic packaging, we compiled the Chemicals in Plastic Packaging Database (CPP-DB), which comprises unique substances with additional substance-specific information such as usage data, physico-chemical properties and uses. The CPP-DB includes plastic monomers, additives, and other substances used during plastics manufacturing, such as solvents and raw materials, and the main non-intentionally added substances (NIAS) such as impurities, reaction by-products or degradation products of e.g. polymers and stabilizers. We ranked the substances in the CPP-DB according to their hazard for human health and the environment, using Classification and Packaging (C&P) hazard categories, and also including endocrine disrupting properties and PBT (persistence, bioaccumulative and toxic) characteristics. Due to the lack of empirical hazard data for many of the substances in the CPP-DB, we also used in-silico tools to bridge data gaps. In this presentation we will use the CPP-DB to present an overview of chemicals associated with plastic packaging, their hazards for human health and the environment, and we will highlight priority hazardous chemicals for substitution. Finally, we will discuss the research needed to allow for a more robust hazard characterization and ranking.

325 A Safe by Design framework to support the development of sustainable nano-enabled products for the restoration of works of art

E. Giubilato, University Ca Foscari of Venice / Department of Environmental Sciences Informatics and Statistics; E. Semenzin, Ca Foscari University of Venice / Department of Environmental Sciences Informatics and Statistics; E. Badetti, University Ca Foscari of Venice / Department of Environmental Sciences Informatics and Statistics; M. Picone, D. Hristozov, University Ca Foscari of Venice / Department of Environmental Sciences Informatics and Statistics; A. Brunelli, University Ca Foscari of Venice / Department of Environmental Sciences Informatics and Statistics; V. Cazzagon, University Ca Foscari of Venice / Department of Environmental Sciences Informatics and Statistics

Innovative nano-enabled products can overcome some issues of the traditional restoration techniques, especially in the case of complex and unstable materials used in contemporary artworks. However, nanomaterials have been demonstrated to be potentially hazardous to both humans and the environment, thus their application for the conservation of cultural heritage requires a proper assessment and management of potential risks. A Safe by Design (SBD) approach can support the early identification and management of uncertainties and risks during an innovation process and allows for the modification of a product design to avoid undesired properties. Within the EU H2020 “NANORESTART” project, a stepwise SBD framework for the sustainability assessment of nano-based products for restoration has been proposed, taking into account the current EU legislative context as well as the specific features of the innovation process in the restoration field. The proposed framework embraces the SBD concept proposed by NANOReg initiative, which uses the Cooper Stage Gate innovation model as backbone to incorporate SBD in structured innovation management processes for nanomaterials. Six steps build up the framework: (a) state-of-the-art assessment, (b) initial formulation, (c) screening hazard assessment (based on CLP self-classification approach for mixtures), (d) advanced hazard assessment (based on the development of an Intelligent Testing Strategy (ITS)), (e) safety assessment (including the definition of Risk Management Measures), and (f) sustainability assessment (where environmental, social, economic and technical criteria are integrated to compare new to conventional products). The framework is focused on application and post-application stages, while the manufacturing stage cannot be included until the industrial up-scale has been finalized. The SBD framework is currently being applied to NANORESTART advanced nano-based formulations for controlled cleaning and surface protection and consolidation. A specific ITS has been defined, including three bioassays: (i) static toxicities, (ii) rapid screening for aquatic toxicity, (iii) a set of tests for cytotoxicity, DNA-damage and mutagenicity. Moreover, specific leaching test protocols are being applied to investigate medium and long-term behaviour of products in post-application stages. The results of the framework application to the most promising formulations will be presented and discussed in detail.

Recent developments in environmental risk assessment for pollinators

326 Managing on the Margins: The confluence of Modern Agriculture and Aiculture

Z. Browning, Browning’s Honey Co., Inc.

In the USA, beekeeping is a hobby, a sideline business, and commercial enterprise. Pollinating our backyard gardens, and local communities is made possible by backyard beekeepers with one hive or more. Beekeepers who aspire to increase their honey production, and crop pollination may have hundreds of hives serving not just their local communities, but their state or regions of the country renting their hives to pollinate specialty crops. Commercial beekeepers migrate with their tens of thousands of colonies to pollinate the nation’s food supply. Once commercial bees have pollinated the majority of specialty crops they head to summer forage areas for a honey crop. The areas of conflict for bees in agriculture encompass the urban backyard garden to the almond orchards of California, pest and pathogens of honey bees are real challenges regardless of location. Habitat loss and pesticide exposure to bees, are greater variables, but no matter what general shared land use is considered for bee hives there are potential conflicts. In each and every case, there are also opportunities to work together with partners and stakeholders for mutually beneficial outcomes. Whether it is water issues, soil contamination, or use issues related to health and safety, there are many cases, pesticide manufacturers must address how the end user interprets the directions for use, and the cultural practices of the products. Regulatory agencies must acknowledge the pesticide end user’s cultural practices of tank mixing pesticides, of fungicide and herbicide impacts upon pollinators, and to combine their agency efforts to protect the entire farm, not just each single crop from each single pest. Sustainable land management strategies for food production or protection of human and animal health from disease vectors must be coordinated to ensure profitable production outputs for all stakeholders. Beekeepers can assist in the development of
scientifically supported risk assessment through participation in research and development. Beekeepers know bees; researchers know research protocols. To understand how honey bees function under migratory beekeeping and crop field conditions beekeepers need to be part of designing the risk assessment research. Beekeepers, no matter the level of beekeeping or number of hives, are eager to be included in research that will help alleviate the risks to honey bees, and native pollinators. Involving beekeepers in risk assessment and research design is key to ensuring the research premise and results truly reflect the real-world of beekeepers and honey bees. In some ideal world beekeepers would be respected for the ecosystem service their honey bees provide to farmers. Beekeeper and farmer would understand their symbiosis in connection with the health of the crop, and the success of the crop’s yield. Both would work to ensure a healthy crop and healthy honey bees to pollinate all crops. One begets the other; each supporting each other: beekeeper and farmer, honey bee and crop (personal examples). As such all stakeholders who rely on honey bees and native pollinators to maintain a healthy ecosystem would balance competing interests to ensure pollinators have clean, plentiful, and diverse forage, pollinators are healthy to provide appropriate pollination services to the ecosystem, and land management is facilitated to reduce soil erosion, protect water, and reduce the threat of disease vectors. (share Bee and Butterfly Fund programs an results)

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A new multi-dimensional method for evidence synthesis and weighting in bee risk assessment
A. Ippolito, R. Sharp, C. Szentes, D. Auteri, EFSA - European Food Safety Authority / Pesticides Unit

In recent years, neonicotinoid substances have often been in the spotlight, particularly due to their effects on bees. Reporting of highly contradictory results catalysed much attention from the scientific community. The great amount of available studies requests approaches able to ensure an effective integration of the available data. To this purpose, EFSA has developed a novel approach for the most recent conclusions on imidacloprid, clothianidin, and thiamethoxam. Risk due to exposure of bees from residues in pollen and nectar of treated crops is used here as a case study to illustrate the methodology. Oral exposure was estimated by combining data on residue levels in pollen and nectar and estimation of bee food consumption. Together with exposure data, higher tier effect data were the weight of evidence exercise. Each endpoint was identified by four dimensions: (I) the magnitude of the observed deviation from the control, (II) the reliability, (III) the level of exposure in the experiment, and (IV) the length of the exposure. In order to visually illustrate these four dimensions of the endpoints and in order to help the interpretation of each line of evidence, a tailored graphical representation was developed. The relevance of each line of evidence was established a priori, based on the relationship with the specific protection goals (SPGs). Integration of the lines of evidence followed a stepwise procedure, giving priority to the higher classes of relevance. Single risk assessment results are beyond the scope of this platform, which aims at communicating the features of this new approach. This was, to our knowledge, the first systematic assessment on such a large body of evidence for this specific topic. The exercise combined systematic reviews and weight or evidence, sharing many aspects with meta-analysis techniques. The approach used in this assessment addressed some issues that commonly undermine the reliability of meta-analysis such as the so-called ‘file-drawer’ problem. Overall, the presented approach ensured significantly more transparency than a fully qualitative expert judgment-driven assessment, but still allowed considering several dimensions in a quantitative way. The need for simplifying the assessment by using fully quantitative measurements that, at present stage, are hardly capable of retaining important qualitative information.

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Pesticide Exposure Assessment Paradigm for Bumble Bees
J van der Steen, Alveus AB Consultancy; C. Cutler, Dalhousie University / Faculty of Agriculture; D. Goulson, School of Life Sciences, University of Sussex; A. Ippolito, R. Sharp, C. Szentes, D. Auteri, EFSA - European Food Safety Authority / Pesticides Unit

The potential exposure routes and actual exposure of the bumble bee queen, workers and larvae are mapped and knowledge gaps are identified. The honey bee is used as the reference point to which the differences in biology, foraging and nursing are compared. Some significant differences in susceptibility to pesticides between Bombus species have also been identified. It is concluded that there are significant gaps in current knowledge for bumble bee species on both realistic levels for some key exposure routes and cumulative exposure that are not accounted for in the current Apis risk assessment protocols.

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Industry research and approaches to improve the bee risk assessment scheme in Europe
E. Pilling, Dow Agrosciences / REGULATORY Sciences; M. Miles, Bayer CropScience UK / Environmental Safety; A. Alix, Dow Agrosciences / Risk Management; J.C. Becker, New York State Dept. of Environmental Conservation / Biology; N. Boulanger, Syngenta Ltd / Environmental Safety; A. Dinter, Chemnitzova Deutschland GmbH & Co. KG / Global Regulatory Sciences; A. Sharples, FMC Agricultural Solutions; G. Weyman, ADAMA; L. Oger, ECPA

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Standardization of an in vitro larval rearing method for stingless bee species Melipona scutellaris for use in toxicological bioassay studies.
A.S. Dorigo, Universidade Estadual Paulista Júlio de Mesquita Filho Unesp Rio Claro / Department of Biology; R. Lopes, Universidade Estadual Paulista Júlio de Mesquita Filho / Department of Biology; S. Rosa-Fonseca, Unesp - Institute of Biology / Departamento De Biologia; C. Cutler, Dalhousie University / Faculty of Agriculture; D. Goulson, School of Life Sciences, University of Sussex; A. Ippolito, R. Sharp, C. Szentes, D. Auteri, EFSA - European Food Safety Authority / Pesticides Unit

In Brazil has the greater diversity of native stingless bees of the world and makes Melipona scutellaris for use in toxicological bioassay studies. Standardization of an in vitro larval rearing method of this species have also been identified. It is concluded that there are significant gaps in current knowledge for bumble bee species on both realistic levels for some key exposure routes and cumulative exposure that are not accounted for in the current Apis risk assessment protocols.

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the progress of the experiments, increasing from 67.1% in the first to 87.8% in the latter, and the morphometric analyses indicated newly emerged workers in vitro with similar sizes to in vivo. The in vitro rearing method described showed a satisfactory survival rate, as well as produced newly emerged workers with similar to those from natural conditions, allowing its use in toxicity tests.

331 Poster spotlight: TU038, TU048, TU052

Understanding human and environmental exposure to chemicals in urban systems

332 Consumption of products - a proxy for changes in chemical flows in urban areas and to the environment?

E. Unedman, D. Bolinius, Stockholm University / Baltic Sea Centre; A. Sobek, Stockholm University / Department of Environmental Science and Analytical Chemistry ACES; M. Löf, Stockholm University / Baltic Sea Centre

Is the threat posed to the environment by harmful chemicals increasing or decreasing? Due to the extremely large number of chemicals and variety of adverse effects, it is challenging to develop indicators for the success of our management of chemical emissions. Indicators for efficiency of chemicals management can be based on a) information on production, trade and use of chemicals, b) emissions, c) concentrations in humans and the environment and c) human and wild-life health, with data on the two latter being most relevant, but also difficult and/or expensive to produce for a wide range of chemicals. In this study, we used estimated consumption of products as point of departure to analyze time trends in use and emissions of chemical substances in the urban society and ultimately in the environment. Data on trade of manufactured products available in Eurostat was combined with chemical composition of products and materials compiled in the Commodity Guide hosted by the Swedish Chemicals Agency. The total mass of manufactured products in the northern Europe decreased slightly between 2003 and 2014. Despite this decline, ca 680 substances with significantly positive time trends due to increasing consumption of many products in which they are likely to be present were identified. We conclude, however, that substantially more data on chemical content of products is needed to successfully use consumption of products as a proxy for changes in chemical flows.

333 High-throughput assessment of use-phase exposures to chemicals in building materials

L. Huang, University of Michigan / Dept of Environmental Health Sciences; V. Nguyen, University of Michigan / Department of Computational Medicine and Bioinformatics; O. Jollet, University of Michigan

Building materials have important contribution to the chemical exposure of the US population. The NHANES data have shown that the blood levels of brominated flame retardants and phthalate plasticizers, tend to be higher in children. The present study thus aims to develop a high-throughput method to determine exposures to chemicals in building materials, which mainly happen during the use phase but are often not considered in traditional LCA. The assessment framework calculates the product intake fraction metric, PiF, to assess consumer exposures during use of products in the building. Based on the building materials Pharos database, 22 product categories for building materials and 218 chemicals were identified. We focus here on 632 unique chemical-product combinations, and assess doses based on PHAROS chemical content data. Chemical emissions from building materials are mainly determined by Dm (diffusion coefficient in building material) and Kma (material-air partition coefficient). Two simple emission models were developed for two types of behaviors: D-limited and K-limited. Use-phase exposures by inhalation, dermal contact, gaseous dermal uptake and dust ingestion were calculated using the PiF metric. Results showed that for SVOCs, the total PiF can be dominated by any of the four exposure pathways. Generally, the chemical-product combinations with low total PiFs are dominated by dust ingestion exposures, while the ones with high total PiFs are dominated by dermal contact exposures. For VOCs, inhalation PiF always dominates the total PiF. Generally, the PiFs of VOCs are similar between 50 days and 15 years, but the daily intake doses during 50 days are much higher than those during the entire 15-year use phase. In contrast, for SVOCs, the emissions and PiFs gradually increase when the duration of use is extended from 50 days to 15 years, but the daily intake doses remain similar over the time. The overall intake dose, which combine the total PiFs and chemical content in building product, can range from 100 to 10^6 µg/kg-d for children. This study demonstrates the approach of high-throughput screening of use-phase exposures for chemicals in building products, which can be further integrated into characterization factors and help improve LCA and Chemical Alternatives Assessment (CAA) of consumer products.

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OPEs - Where do they come from, where do they go? A case study from Toronto, Canada

T.F. Rodgers, University of Toronto / Chemical Engineering and Applied Chemistry; Y. Adjei-Kyereme, University of Toronto / Earth Sciences; C. Yang, University of Toronto / Department of Earth Sciences; S. Harris, Cancer Care Ontario, University of Toronto / Population Health and Prevention, Prevention and Cancer Control; L. Jantunen, Environment and Climate Change Canada; M.L. Diamant, University of Toronto / Department of Earth Sciences

Organophosphate Esters (OPEs) are used in a wide range of consumer products and building materials resulting in their ubiquitous presence in the indoor and outdoor environment. Concentrations of OPEs in indoor air can be relatively high, in the range of 10’s ng/m3 in air. Concentrations are also relatively high in urban media (e.g., low ng/L levels in urban surface waters) and OPEs are commonly found in remote Arctic air. We hypothesize that elevated levels of OPEs in the indoor environment are a source to the outdoor urban environment and then to surrounding regions. To test this hypothesis, we estimated emissions of OPEs to indoor air followed by release to outdoor air, and then compared these “bottom-up” emission estimates to “top-down” estimated aggregate emissions to outdoor urban air. We used the approach of “inverse modelling”, whereby emissions are back-calculated from measured air concentration. “Bottom up” estimates were used estimating the Multimedia Indoor Model that has been used to calculate emissions of PBDEs and PCBs to indoor air, based on indoor air concentrations measured in a study of 51 homes in Ontario, Canada. “Top down” emissions were calculated using the Multimedia Urban Model based on measured Toronto outdoor air concentrations, main receptors for these molecules. Pharmaceuticals can be referred to the indoor and outdoor environment from multiple point sources within a city and can then dissipate as they move away from the point of release. Spatially referenced hydrological models such as LFS2000-WQX, GREAT-ER and Ph/AE have been developed to address this issue. Many pharmaceutical monitoring studies have indicated that temporal significant fluctuations exist in rivers receiving urban wastewater. Currently, spatial models typically overwork the temporal aspect of pharmaceutical concentrations by predicting an annual average concentration usually based on conservative flow estimates. The relevance of these estimates in comparison to real world temporal variations in pharmaceutical exposure has yet to be determined. In this study we attempt to quantify and understand the drivers behind spatiotemporal fluctuations in the concentrations of pharmaceuticals in a city. Monitoring data for 33 pharmaceuticals were obtained monthly at 11 sites in two rivers that run through the City of York, UK. This data was compared with local monthly prescription, tourism and flow data. In the smaller River Foss, a strong relationship was found between measured concentrations and prescription amounts divided by flow. This trend was not replicated in the larger more urbanised River Ouse. Analysis of pharmaceutical loads indicate that seasonal differences exist in the Ouse, but not in the Foss. Seasonal variability in WWTP removal efficiency is expected to be a factor. In-stream losses of up to 75% were found in the River Foss during summer months and all pharmaceuticals studied followed a sinusoidal loss pattern through time. Significant losses were not identified in the River Ouse. Finally a risk assessment indicated that risk quotient ratios can vary over 3 orders of magnitude (paracetamol) throughout the year and that direct emissions of OPEs to outdoor air, such as OPEs, are transiently. Identification of exposure drivers at this unparalleled spatiotemporal scale provides important information that may help improve the accuracy of exposure models and help ensure risks are not overlooked.

336 Past vs. recent emissions of PCBs from the city of Brescia (Italy): coupling monitoring data with a multimedia fate model to investigate PCB regional fate

E. Terzaoui, University of Insurbria (Como) / Department of Science and High Technology, Como; M. Morselli, University of Insurbria / Department of Science

Drivers of pharmaceutical exposure in urban river systems

E.E. Burns, University of York / Chemistry; L. Carter, University of York / Environment Department; J. Thomas-Oates, University of York / Chemistry Department; A. Boxall, University of York / Environment Department

Pharmaceuticals are heavily used by society and urban environments are one of the main receptors for these molecules. Pharmaceuticals can be released to the indoor and outdoor environment from multiple point sources within a city and can then dissipate as they move away from the point of release. Spatially referenced hydrological models such as LFS2000-WQX, GREAT-ER and Ph/AE have been developed to address this issue. Many pharmaceutical monitoring studies have indicated that temporal significant significant fluctuations exist in rivers receiving urban wastewater. Currently, spatial models typically overwork the temporal aspect of pharmaceutical concentrations by predicting an annual average concentration usually based on conservative flow estimates. The relevance of these estimates in comparison to real world temporal variations in pharmaceutical exposure has yet to be determined. In this study we attempt to quantify and understand the drivers behind spatiotemporal fluctuations in the concentrations of pharmaceuticals in a city. Monitoring data for 33 pharmaceuticals were obtained monthly at 11 sites in two rivers that run through the City of York, UK. This data was compared with local monthly prescription, tourism and flow data. In the smaller River Foss, a strong relationship was found between measured concentrations and prescription amounts divided by flow. This trend was not replicated in the larger more urbanised River Ouse. Analysis of pharmaceutical loads indicate that seasonal differences exist in the Ouse, but not in the Foss. Seasonal variability in WWTP removal efficiency is expected to be a factor. In-stream losses of up to 75% were found in the River Foss during summer months and all pharmaceuticals studied followed a sinusoidal loss pattern through time. Significant losses were not identified in the River Ouse. Finally a risk assessment indicated that risk quotient ratios can vary over 3 orders of magnitude (paracetamol) throughout the year and that direct emissions of OPEs to outdoor air, such as OPEs, are transiently. Identification of exposure drivers at this unparalleled spatiotemporal scale provides important information that may help improve the accuracy of exposure models and help ensure risks are not overlooked.
and High Technology; G. Raspa, Sapienza University of Rome / Department of Chemical Engineering Materials and Environment; A. Di Guardo, University of Insubria/Department of Science and High Technology

Different monitoring campaigns showed higher PCB concentrations in the air of cities compared to rural areas, indicating the presence of ongoing emission sources in urban locations. This is the result of both the past usage of these chemicals in building materials and electrical equipment from which they can volatilise and/or leach into the e-waste composted soils. It was estimated that cities could emit up to about 1 tons/year of PCBs (Gasic et al., 2009; Diamond et al., 2010), which can be transported to rural sites, posing adverse effects to human and ecosystems. The city of Brescia, is characterized by the presence of a factory (Caffaro s.p.a.) that produced PCBs between 1930-1984 and its surrounding areas were found to be heavily contaminated with high PCB concentrations in soil at mg/kg levels. For this reason, about 200 ha of this city were declared National Priority Site for remediation (SIN Brescia Caffaro) by the Italian authorities, representing an important secondary source of PCBs to the atmosphere.

The aim of the present study was to investigate the potential of the Brescia city in driving the PCB contamination at regional scale up to 100 km from the point source and the current effects on air concentrations, combining measured data and a multimedia mass balance model. Different sampling campaigns were organized to collect samples of soil and leaves along four 100 km transects that ran in NW, NE, SW and SE directions considering the production plant as starting point. In each sample, the following PCB congeners were determined: PCB 28, PCB 52, PCB 101, PCB 153, PCB 138, PCB 180 and PCB 209. The results were analysed to understand the relative contribution of far- and near-field routes, determining the temporal PCB concentrations with distance from the point source. Furthermore, a spatially resolved version of a dynamic air-vegetation-soil model (SoilPlusVeg model) was used to 1) predict a temporal emission profile from the city, 2) verify if an emission source strength previously predicted for this city justifies soil concentrations in the surrounding area and, 3) evaluate the importance of other sources and processes involved in the complicated PCB transport and fate in Brescia. This study shows how a combined modeling approach could be used to unravel the past and recent impact of PCB emissions from a source on the surrounding areas at a regional scale.

337 Using a Dynamic Aggregate Exposure Model to Identify Far- and Near-Field Contributions to Human PCB Exposure through Time

L. Li; University of Toronto at Scarborough / Department of Environmental Sciences; J.A. Arnot, ARC Arnot Research & Consulting / Adjunct Professor, Department of Physical & Environmental Science/Department of Pharmacology and Toxicology; F. Wania, University of Toronto at Scarborough / Physical and Environmental Sciences

Humans are exposed to polychlorinated biphenyls (PCBs) through “far-field” sources from the ingestion of contaminated food from aquatic and terrestrial environments and “near-field” sources from building materials in indoor environments. Earlier models that consider far-field exposure only tend to underestimate historical body burdens. In this presentation, we will explore the time-dependence of the relative contribution of far- and near-field routes to PCB aggregate human exposure to PCBs, to explain the discrepancy between previous far-field only model predictions and observations. We develop a mechanistic model that incorporates dynamic substance flow analysis, indoor-urban-rural fate modeling, and bioaccumulation and human toxicokinetic modeling, enabling a dynamic and mechanistic description of the complicated continuum from annual industrially processed amount of PCBs to the human uptake rate. The model is applied to simulate the time-variant exposure of Swedish women to PCB-28 and PCB-153 from 1930 to 2030. In terms of a female’s lifetime longitudinal exposure, our modeling indicates that route-specific contributions to aggregate human exposure change with age and differ among birth cohorts: Near-field exposures are notable during childhood and teenage years, as well as for female cohorts born earlier, when the indoor environment was more highly contaminated. In terms of the exposure of individuals of different ages at the same time (i.e., the cross-sectional age-exposure relationship), the dominance of far- or near-field routes differs little among ages, but is largely dependent on the time a cross section is “sampled”: Near-field routes dominate in the past (e.g., the year 1956) whereas far-field routes become predominant more recently (e.g., 1986 and 2016). This finding suggests that the dearth of PCB biomonitoring studies before 1990s has also contributed to the general belief that far-field sources dominate human PCB exposure. It further implies that there may also be a similar shift from a near-field dominance to a far-field dominance among a wide range of currently-used indoor chemicals, such as flame retardants. This work improves our understanding of the exposure dynamics, which would be beneficial for future pertinent management actions for exposure reduction and prevention.

Microplastics in freshwater and terrestrial systems - fate, monitoring and biological interactions (III)

338 Modelling of the environmental release of macro- and microplastics for seven different polymers

D. Wengier, Empa Swiss Federal Laboratories for Materials Science and Technology; B. Nowack, EMPA

Microplastic exposure is a burning topic in environmental research, but few large scale exposure studies have yet been performed in freshwaters. Assessing the emissions of plastic is possible using a life-cycle oriented approach, and permits to compare the flow magnitudes for different sources. With our ongoing study, we aim at providing large scale predictions of macroplastic and microplastic exposure in Europe using Probabilistic Material Flow Analysis (PMFA). The environmental flows of seven different commodity thermoplastics are estimated based on societal data. The polymers are chosen for their popularity of use and the frequency at which they are reported in the environment: low-density polyethylene (LDPE), high-density polyethylene, polypropylene (PP), polystyrene (PS), expanded polyurethane foam (RPF), polyvinyl chloride (PVC) and polyethylene terephthalate (PET). The probabilistic aspect of the PMFA framework permits one to account for the various uncertainty sources and give a quantitative estimate of the final confidence in the results. In a first step, the anthropogenic life cycle of these seven polymers is modelled, from production to end of life of a total of 35 product categories. Various trade flows are included, as well as the life cycle of textile applications. This enables us to present an accurate estimation of the European and Swiss consumptions. In a second step, every stage of the life cycle is analyzed and the voluntary or inadvertent emissions are assessed. The emission-specific pathways between the anthroposphere and the environment are described, and the resulting environmental flows are compared. Modelling the emission pathways between prime release and final discharge enables to pin-point the principal plastic pollutants sources and the possibilities for pollution mitigation.

339 Modelling Microplastics in Rivers in the US

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Pollution with nano- and microplastics (MPs; particles < 5 mm) is a topic of emerging concern and as such receives growing interest. Although measurement and monitoring data are indispensable, there also is a need for estimated concentrations to enable prospective assessments and to guide analysis of retrospective ecological analyses. Besseling et al (2017) provided the NanoDUFLOW model, a detailed MPs generation-sedimentation model integrated in a hydrological and particle transport model. A much larger scale model potentially suitable to simulate MPs originating from WWTPs is the iSTREEM® model, which has been developed to estimate chemical concentration distributions for all rivers and streams of the USA receiving WWTP discharges. Here we merge these two riverine modeling worlds: NanoDUFLOW with iSTREEM for MPs, to simulate the loading of MPs from WWTP point sources in US rivers and to assess export to the Great Lakes for a range of particle sizes. This combines the mechanistic realism of NanoDUFLOW, accounting for formation and settling of heteroaggregates, with the US well-established iSTREEM implementation. We modeled floating as well as non-float MP, for diverse sizes, from 100 nm to 10 mm, a range that incorporates the theoretical parabolic size-setting relationship reported by Besseling et al (2017). Depth dependent in-stream first order removal rate constants simulated with NanoDUFLOW were combined with standard iSTREEM® output (which was used to simulate the emission, transport and water column concentrations of MP) in an Excel-based post-processing phase, without modifying the iSTREEM® model directly. Simulations were spatially explicit with MP concentrations being modeled for the Sandusky River watershed in Ohio (~100 km) and the Great Lakes. Emissions were used as input with equal concentration over time for each of the 20 WWTPs within the watershed. Modelling results show the effects of population density, MP size and density on riverine concentrations and export to Lake Erie. Buoys as well as the smallest non-float MP fractions can be transported over long distances, reaching receiving waters such as the Great Lakes. In contrast, larger non-float MP settle more locally in the vicinity of the WWTPs. Simulating depth-dependent removal as demonstrated here could be incorporated into the core iSTREEM® code in order to efficiently process all US waterways impacted by WWTPs, as well as examining ultimate marine discharge proportions by particle size. 

340 The routes to uptake and bioaccumulation of nanoplastics in freshwater sediments

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Plastic contamination of freshwater sediments is well documented, and whilst quantitative measurement of routes of tissue and human uptake is important, quantifying their potential to transfer through food chains can contribute towards the data required to inform decisions as to the suitability of classifying nanoplastics as POPs. This study provides initial insights to address this question in...
an ecologically relevant system, using the freshwater aquatic worm Lumbriculus variegatus, representing an entry point for nanoparticles from abiotic compartments of sediments, into biota. The role of surface functionalisation of fluorescently dyed nano-polystyrene (50 nm) upon their uptake is systematically examined using a combination of techniques including a novel fluorescence assay and fluorescence microscopy. A series of exposure scenarios are used to test the efficacy of different routes of uptake into the worm. Associations of nanoparticles to the worms’ surface are examined in waterborne exposures, whilst dietary uptake is tested using nanoparticles associated with an algae food source. The accumulation of nanoparticles directly from contaminated sediments is also investigated, alongside the fate of these particles in sediments to assess the relationship between nanoplastic mobility and accumulation. Results indicate that pristine nanoplastics and contaminant-exposed nanoplastics both with and without degradation show uptake in a similar manner. Dietary uptake of a nanoplastic associated algal food source, with carboxylated and aminated plastics experiencing greater uptake than non-functionalised particles.

431 Life-history and biochemical responses of Chironomus riparius exposed to different sized microplastics

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Freshwater basins are an integral part of microplastics life-cycle, being a repository of plastic micro-debris. In fact, the levels found so far are similar to those found in marine environment and shoreline regions. The deposition and persistence of plastic micro-debris in sediments (lakeshores and riverbanks) makes them a long-term available for benthic species. The objective of this study was to investigate developmental, and physiological effects induced by the presence of polyethylene nanoparticle (PE) in Chironomus riparius, due to its key-role in the freshwater ecosystem. For that partial life cycle tests using different sized polyethylene particles (PE 40–48 µm; PE 125 µm and PE 350 µm) allowed. Evaluation of effects on C. riparius larval growth and emergence patterns while acute exposures were used to assess effects in parameters related to neurotransmission (ACHE); antioxidant defences and biotransformation (CAT; GST total glutathione levels); oxidative damage (LPO); cellular energy allocation (CFA) and immune response (phospholipase). Exposure to PE 40–48 µm caused deleterious effects at lower concentrations in comparison with larger particles in several parameters: larval growth and development time of both male and female imagos and on emergence rate. PE 40–48 µm were then selected to assess effects on physiological homeostasis. Acute exposures to PE 40–48 µm generated alterations in C. riparius larvae antioxidant and biotransformation enzymes activities (CAT; GST and total glutathione) and activation of immune response (induction of phenoloxidase). Larvae exposed to microplastics showed also decreased energy reserves. Our study highlights the potential deleterious effect of microplastics for aquatic invertebrate populations. Results will be discussed in terms of effects of different sized plastic particles on different levels of biological organisation within freshwater invertebrates and on the needed and ongoing research aiming to address the long term and indirect effects of these particles for natural populations and ecosystem functioning.

432 The effects of rigid and flexible Polyvinyl chloride (PVC) microplastic particles on the transcriptome of Daphnia magna

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Microplastics are ubiquitous in aquatic ecosystems, posing as a threat to biota of all trophic levels, as they have the potential to leach out incorporated additives, such as plasticizers, to the surrounding medium. Yet the question arises, if possible effects are based on the polymer type alone, or if incorporated additives are regulated and 4 down regulated) related to the GO functionalised particles.

When ecotoxicology meets trophic ecology

433 Poster spotlight: TU149, TU150, TU151

Does stress propagate along aquatic food chains? An experimental approach with a tri-trophic brown food chain

E.L. Fernandes, University of Koblenz Landau; M. Bundschuh, Swedish University of Agricultural Sciences / Department of Aquatic Sciences and Assessment; R. Schulz, University of Koblenz-Landau / Institute for Environmental Sciences; R. Schaefer, University Koblenz Landau / Institute for Environmental Sciences Pollution is a major driver of ecosystem change resulting in alterations in food webs and therefore ecosystem processes. Some pollutants such as endocrine and insecticides are taken up by terrestrial plants and may enter aquatic systems with plant parts such as leaves that are an important energy source in stream food webs. Therefore, food web interactions across the aquatic-terrestrial boundary may be affected through alterations in food quality. Here we studied the effects of a systemic insecticide, the neonicotinoid imidacloprid, and their potential propagation in a brown food chain. The model food chain consisted of imidacloprid contaminated terrestrial leaves (alder, Alnus glutinosa Gaertn.), aquatic meromictic invertebrate decomposers (Protonemura sp.) and predators (Isoperla sp.). Effects of imidacloprid on survival and growth of decomposers and its leaf processing were assessed in a mesocosm setting. Therefore, decomposers fed on control or contaminated leaves for 3 days on the microcosms. Every 6 hours the number of dead individuals was recorded. Potential propagation of imidacloprid effects were assessed by transferring surviving decomposers to cages containing the predator. The cages were deployed in an unpolluted stream for 9 days after which predators’ growth was analysed. Imidacloprid concentrations increased within the contaminated microcosms over time. The presence of imidacloprid in the water was associated with lower survival rates of decomposers and leaf decomposition. The effects can propagate through food chains and result in indirect effects in predators. Future studies should elucidate the spatiotemporal dynamics of exposure and uptake given that imidacloprid leaks from plant material and may influence downstream food webs directly and indirectly.

435 Accounting for trophic relationships in fish bioconcentration models applied with emergent-pollutants risk-assessment tools

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In the context of the SOLUTIONS EU FP7 project, we applied non-steady state bioconcentration models to predict concentrations of organic compounds in fish. A foodweb perspective was taken, accounting for uptake from water as well as from food, and accounting for different trophic relationships for several fish species used for human consumption. The foodweb bioconcentration model will be applied for a large number of emerging pollutants and a large number of locations (around 25,000 sub-catchments in the major European catchments). Water concentrations at these locations are obtained from chemical fate modelling using the STREAM-EU model. As a case study, results for 24 WFD priority substances are presented here. Predicted concentrations will be input to human health risk assessment. The model also provides insight in how trophic relationships together with species and compound characteristics determine bioconcentration and thus ecotoxicological risk. The core of the foodweb model is a bioconcentration model for neutral and ionisable organic compounds (Arnot & Gobas 2004; Armitage et al. 2013) underlying each fish component. It calculates for given environmental conditions (pH, temperature) the uptake and elimination rates defining the one-compartment model of the internal concentration dynamics. The considered foodweb contains fish components with different trophic relationships, representing fish species used for human consumption with different body size and lipid content, chosen to represent extreme cases with respect to expected bioconcentration. Internal concentrations in phyto- and zooplankton are assumed to be in instantaneous equilibrium with water concentration. For 24 WFD priority substances concentration time-series per sub-catchment from the STREAM-EU model were used as input to the foodweb bioconcentration model. Results were summarized in monthly and annual maximum and mean concentrations for all foodweb components in each sub-catchment and displayed in maps covering the
EU. From these results, e.g., median concentrations can be calculated per catchment or over all catchments. Concentrations in fish depend on local exposure pattern and differ per subcatchment. They also depend on trophic position in a compound-specific way: any of the three fish components can be worst-case. To indicate risk to human health, concentrations need to be related to standards of e.g., acceptable daily intake.

346 Model-based explorations of the variability in lake trout BAFs caused by physiology and trophic relationships

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Because dietary uptake of fish is often a major vector of human exposure to persistent organic pollutants (POPs), much effort is directed towards a quantitative understanding of fish bioaccumulation with the help of mechanistic models. Such models require the input of the growth, feeding and respiration rates of a fish. However, often little consideration is given to the interdependency of these physiological parameters. Here, we calculate the bioaccumulation factor (BAF) of hypothetical POPs, with log $K_{ow}$ values ranging from 4.5 to 8.5, in lake trout (Salvelinus namaycush), with a food web bioaccumulation model that uses bioenergetic equations to ensure that the physiological parameters applied to a species are internally consistent (i.e. energetically balanced). Empirical growth rates and diets for lake trout were used to estimate a carbon productivity (C) and nitrogen productivity (N) of the lake trout based on the metabolic rates of the living organisms. Nutrient concentrations in the lake trout depend on the nutrient content of the aquatic and terrestrial prey organisms. We found less aquatic and terrestrial prey organisms in the co-occurrence of pesticide use, habitat degradation and excessive nutrients, which are low in the food web. Moreover, very large trout in Great Slave Lake with higher energy requirements feeding on an energy rich diet have lower BAFs compared to the same sized trout in Lake Memphremagog feeding on less lipid rich rainbow smelt.

347 Influence of an agriculture-associated toxicity gradient on a riparian predator-prey relationship in Romania

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Riparian systems are complex landscapes with complex nutrient cycles and interactions between aquatic and terrestrial ecosystems. The toxicity gradient was negatively related to the number of individuals of spiders and the number of species. Fish communities in water and on land, resulting in complex response patterns of aquatic-terrestrial predator prey relationships. Therefore complex response patterns may arise in terrestrial predators feeding amongst others on aquatic prey. While agricultural landscapes in most European countries have been intensified, resulting in the co-occurrence of pesticide use, habitat degradation and excessive nutrients, traditional low-intensity agriculture can still be found in Central Romania. We investigated the potential effects of land use related stressors including pesticides on aquatic-terrestrial predator-prey relationships using stable isotope analysis. We sampled spider communities and measured their intake of aquatic prey in 19 riparian areas around Cluj-Napoca, Romania. To investigate the spiders' diet, aquatic and terrestrial prey organisms were caught. We collected the orbweb Tetragenatha sp. and the ground-dwelling spider Pardosa sp. to analyse their stable carbon and nitrogen signals. Nutrient concentrations in the stream were slightly positively associated with the proportion of aquatic prey of Pardosa sp. This may be explained by nutrients in the stream increasing productivity of primary producers and invertebrates, resulting in a larger biomass of emerging insects. The toxicity gradient was negatively related to the number of individuals of spiders and the number of species. Although we found clear differences in the proportion of consumed aquatic prey of spiders, the proportion of it was not related to the toxicity gradient. Thus, potential effects of pesticides in the aquatic system did not affect the proportion of consumed aquatic prey organisms of riparian spiders. We found less individuals of Tetragenatha sp. when they consumed more aquatic prey. This might be due to an accumulation of toxicants in the spiders or a higher competition between the species due to resource shortage. Riparian spiders can be affected directly by agricultural land use but also indirectly via prey consumption. Changes in riparian spider communities and their diets are presumably driven by multiple stressors.

348 Migration effects on pollutants in eggs of Arctic-breeding gese

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Arctic breeding barnacle geese (Branta leucopsis) are a herbivorous species that migrate from the United Kingdom to the Arctic every summer to breed. Females utilise resources towards reproduction along the flyway, including distant resources (United Kingdom and Northern Norway) and local resources (Svalbard) relative to the breeding grounds. Depending on migration route, allocation of resources towards egg production may differ within a breeding goose population. Thus different energy sources may also affect how pollutants are taken up and deposited to eggs, including those which are both protein and lipid soluble. In order to examine the effect of migration on pollutants in eggs, a field study was carried out during the breeding season of 2016. Eggs (N = 60) were collected at an island breeding colony in Svalbard and several hundred grams of vegetation (N = 15 sample collections) was collected at different sites along the goose's flyway. Results of ringed geese also took place in Northern Norway. Egg and vegetation samples were analysed for stable isotope of carbon (δ^{13}C) and nitrogen (δ^{15}N), as well as pollutants including protein-associated poly- and perfluoralkyl substances (PFASs), lipid soluble polychlorinated biphenyls, and hexachlorobenzene (PCBs and HCB). Stable isotope ratios in eggs could not be explained by POPs, but emerged to POPs-like perfluorinated compounds due to the same pollution signal, but stable isotopes of nitrogen appeared to be fuelled by distant resources in United Kingdom and Northern Norway. When examining pollutants individually, there was no relationship found between stable isotopes and pollutant concentrations. However, when combining pollutants together as part of a multivariate analysis, it was found that egg laying date contributed to the variation in PFAS levels across eggs, whereas age for PCBs. Protein associated pollutants (PFASs) may be more influenced by migration strategy than lipid soluble contaminants (PCBs and HCB), due to protein stores being a more limiting energy source during migration than lipids. This knowledge furthers our understanding on how pollutants operate within Arctic terrestrial ecosystems, and the interaction between climate and pollutant bioaccumulation in highly seasonal environments.

349 Trophic Magnification of Persistent Organic Pollutants Within A Terrestrial Food-Web of An Avian Top Predator, the Cooper's Hawk (Accipiter cooperii)

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Several types of legacy persistent organic pollutants (POPs), such as PCBs and DDTs, but not for PCBs. Protein associated pollutants (PFASs) may be more influenced by migration strategy than lipid soluble contaminants (PCBs and HCB), due to protein stores being a more limiting energy source during migration than lipids. This knowledge furthers our understanding on how pollutants operate within Arctic terrestrial ecosystems, and the interaction between climate and pollutant bioaccumulation in highly seasonal environments.

350 Integrated approaches in ecotoxicology: bridging the gap between experimental toxicology and mechanistic modelling
Toxicokinetic-toxicodynamic models as new tools for environmental risk assessment

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Today, the Environmental Risk Assessment (ERA) for chemicals is based on fitting standard dose-response (DR) models to quantitative data. Such data are mostly collected from standard toxicity tests, from which the concentration leading to 50% lethality or effect (LC50 or EC50) is usually estimated at the end of the exposure. In this form of evaluation, the fact that endpoints are monitored over time is not fully exploited. Standard DR models do also assume that the exposure concentration remains constant during the experiment, what makes it difficult to extrapolate the results to more realistic time-varying exposure profiles. To overcome this gap at the organism level, the use of the toxicokinetics-toxicodynamics (TKTD) models is suggested, because TKTD models describe the effects of a substance by integrating the dynamics of exposure [1]. Indeed, TKTD models have many advantages in terms of mechanistic understanding of the chemical mode of action, of deriving time-independent parameters, of interpreting time-varying exposure and of making predictions for untested and time-variable exposure. Another advantage of TKTD models for ERA is that they make it possible to calculate any LC50 or EC50, for arbitrary effect strength r and any given exposure duration t. Nevertheless, being based on differential equations their mathematical complexity makes it necessary to numerically integrate the equations when fitting the model to data, so that in practice TKTD models are still not used, there are no data to allow us to use this type of models for regulatory risk assessment without suffering any technicalities, the availability of a software environment for an easy handling of TKTD models would be of great value. That is the aim of the R package ‘mosa’ in its new version 3.0.0. [2] In this presentation, we will give an overview of TKTD models with a focus on the General Unified Threshold model for Survival (GUTS) [3]. Handling GUTS models is a hands-on session that will be illustrated with one example dataset. Finally, the added-value of TKTD models for ERA will be discussed based on a number of different datasets.

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Lethal and sublethal impacts of neonicotinoids and copper nano-particles on the energy budgets of an estuarine amphipod

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Estuaries are major recipients of run-off pesticides from agricultural and urban origin, including neonicotinoids and nano-based copper formulations. Neonicotinoids have rapidly become the most widely used insecticides globally, and have been implicated for harming pollinators and non-target species at levels below existing US EPA toxicity thresholds. With most research conducted on isolated effects, we are lacking data to allow us to use these models for regulatory risk assessment without suffering any technicalities, the availability of a software environment for an easy handling of TKTD models would be of great value. That is the aim of the R package ‘mosa’ in its new version 3.0.0. [2] In this presentation, we will give an overview of TKTD models with a focus on the General Unified Threshold model for Survival (GUTS) [3]. Handling GUTS models is a hands-on session that will be illustrated with one example dataset. Finally, the added-value of TKTD models for ERA will be discussed based on a number of different datasets.

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A biology-based model to analyze growth data of earthworms exposed to copper at different development stages

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Assessing effects of a contamination on populations requires getting data on the whole life cycle and accounting for sensitivity of sensitivity throughout this life cycle. Copper contamination is one of the greatest concerns in agricultural soils, especially in vineyards. This contamination may have negative effects on soil fauna, such as earthworms, which are recognized as ecosystem engineers providing very valuable ecosystemic services. To assess effects of copper on earthworm populations, we tested the impact of a commercial formulation of copper fungicide, i.e., Cupra Micro® (50% copper oxichloride), on different endpoints, including growth, for the earthworm species Aporrectodea caliginosa, one of the dominant earthworm species in agricultural fields. We performed original tests assessing the impact of copper on the growth at 3 different development stages with many times of repeated measurements: new-born juveniles (10-15 mg), small juveniles (90-110 mg) and large juveniles (260-340 mg). To analyze simultaneously all the data, we developed and used a biology-based model. This model is based on the DEB (Dynamic Energy Budgets) theory. A DEB model was set up and validated under controlled conditions, using different food conditions. The growth data were then analyzed with a toxicodynamic model (taking into account the effect of metal concentrations on growth and differences between development stages) coupled with a DEB-based toxicodynamic model. Our results showed a drastic inhibition of growth once a No Effect Concentration, estimated at 65 mg kg-1 of copper for all stages, was exceeded. The time-profile of the effects was fully accounted for by the model. In conclusion, our model provided a relevant analysis of the toxicity data and provided a tool to understand the mechanisms of copper toxicity to earthworm growth throughout development. The next step of our research will be to combine our biology-based model and reproduction data in a dynamic population model.

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Connecting suborganismal and organismal responses using Dynamic Energy Budget Modeling and the ecological model species Fundulus heteroclitus exposed to dioxin-like chemicals

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Comprehensive and efficient management of ecological risk depends on our ability to quantitatively extrapolate the effects of stressors across levels of biological organization. Adverse Outcome Pathways (AOPs) connect sub-organismal mechanistic molecular data to organismal outcomes, while bioenergetics models, such as Dynamic Energy Budget (DEB) can extrapolate from individual to ecological-level effects of toxicant exposure. However, the connection between the two modeling frameworks remains a challenge. The molecular mechanisms underlying Key Event (KE) relationships defined in AOPs are often poorly known, even for well-studied compounds; thus, the mechanistic linkages between KEs and effects on DEB processes are often difficult to discern. Further, AOPs whose adverse outcome is lethal to the individual, such as sublethal PCB126 exposure leads to slower growth in larval killifish (Nacci unpublished data). Therefore, this system offers a framework to test our ability to connect effects observed at the suborganismal level to bioenergetic processes through AOP and DEB modeling. We will measure suborganismal effects of DLCs (cytochrome P450 activation & transcription) along with effects on development, growth, and reproduction. We will give an overview of our objectives and methods and report preliminary findings fitting DEB models and predictions of the effect of DLCs on the bioenergetics of killifish.

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Quantitative Adverse Outcome Pathway Modelling of Endocrine Active Toxicants in Rainbow Trout

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We are investigating best approaches to utilizing in vitro activation data for estimating impacts on reproduction in trout and salmon. Our goal is to facilitate development of quantitative in vitro – in vivo extrapolation (IVIVE) methods to support adverse outcome pathway (AOP) based toxicity testing. We tested a diverse suite of toxicants using cellular assays based on the female rainbow trout pituitary, ovary and liver. Each assay measures an essential reproductive endocrine function...
such as estrogen synthesis and secretion by ovarian follicles. In vitro results are extrapolated to metrics of reproductive performance (fecundity, fertility, egg size) in trout using a mathematical model of the trout hypothalamic-pituitary-ovary-liver (HPOL) axis. We evaluated IVIVE by comparing predicted effects against laboratory results obtained from a yearlong exposure of female trout to four different chemicals: tamoxifen (bistomerized into the anti-estrogen 4-OH-tamoxifen), prochloraz (interferes with oocyte maturation), fluoxetine (SSRI pharmaceutical largely negated in our in vitro assays) and trenbolone (potent synthetic androgen). A single water exposure level was tested for each chemical, guided by preliminary studies and a desire to use a maximum tolerated exposure that still allowed spawning to occur. Laboratory exposures began 10 d after the first spawning cycle and lasted until time of ovulation and completion of the second spawning cycle 12-14 months later. Trout were euthanized and total fecundity determined along with egg mass and diameter, fertility, hatching success and larval growth. Results indicated no effect on fecundity was observed except after the 60 ng/L trenbolone exposure, which caused regression of ovarian growth and a failure to spawn in all exposed fish. Fluoxetine had no effect on reproduction. The most significant effects on egg quality occurred after the 500 ng/L tamoxifen (30% decrease in egg mass and diameter) and 20,000 ng/L prochloraz (increase in atretic/fertile eggs) exposures. The tamoxifen induced decrease in egg size translated to significantly smaller larval at 20 dph. The HPOL model, guided by in vivo testing, accurately predicted the reproductive effects of prochloraz and tamoxifen and the lack of effect by fluoxetine. Our results support the use of biologically based mathematical models of physiological systems in AOP testing. Supported by EPA-STAR grant R835167.

355 Development of a PBPK model for metal accumulation in fish infected with acanthocephalan parasites

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Fish are affected by both exposure to metals and pathogen. Each of these stressors might have effects on the response of fish to the other. Some of the efforts have recently been in developing kinetic models for predicting metal accumulation in fish-parasite systems. Our previous model allows for investigating the relationship between the accumulation in the whole fish and in the acanthocephalan, but does not include the mechanisms how metals are accumulated in parasites. Physiologically based pharmacokinetic (PBPK) model has been used for simulating the non-specific accumulation of pollutants. However, the capability of this model for simulating fish-parasite systems has not been investigated. We developed a PBPK model for simulating Ag accumulation in the host-parasite system: chub (Squalius cephalus) and the acanthocephalan Pomphorhynchus tetracollicis. The acanthocephalan was considered a compartment, similar to blood, storage, gills, kidney, liver, and intestine. Metal accumulation in the system was modelled as a function of internal (i.e. exchange between different compartments) and external (i.e. exchange with water) factors. The transport from blood to other compartments depends on the diffusive exchange and the fraction of metals dissolved in blood plasma and was assumed to be independent of the infection state. The rate constants for this transport were parameterised based on published data. The model was then calibrated by MATLAB-based AMIGO modelling software for determining the rate constant for Scioptia from storage, gills, kidney, liver, and intestine to blood as well as the external exchange. Model calibration was carried out by using experimental data generated when the infected chub were exposed to Ag in 48-day exposure and 51-day depuration periods. The initial results from model calibration show potential of the PBPK model for simulating the accumulation of metals in fish-parasite systems. For example, the model could simulate the changes in the concentration of Ag in storage, gills, and intestine. The stability in the concentration of Ag in kidney was also simulated by the model. However, the model is being further calibrated to improve its capacity for modelling the accumulation in liver and in the acanthocephalan. The observed weakness of the current version in modelling the accumulation of Ag in these compartments might be related to the approaches for simulating the excretion of organo-metal complexes to intestine.

Solutions for emerging pollutants - Towards a holistic chemical quality status assessment in European freshwater resources (III)

356 High-throughput exposure and risk modelling of chemicals in European river basins

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SOLUTIONS is a EU 7th Framework Programme R&D project that aims at strengthening the management of emerging contaminants. It developed a collection of integrated models (the “Model Train”), to increase our understanding of issues related to emerging chemicals in Europe’s river basins, to support the prioritisation of chemicals and the abatement of the problems they cause and to evaluate future scenarios. The model train consists of four key building blocks: (i) the prediction of substance properties based on their molecular structure, (ii) the simulation of emissions, (iii) the simulation of fate & transport, and (iv) the characterisation of the risk of mixtures of chemicals for human health and aquatic ecosystems. The Model Train does not rely on extensive substance-specific input data. This implies that all the required data are truly “emerging” chemicals and for large numbers of chemicals (“real world exposure scenario”). The approach is validated for well-studied substances and data-rich basins. On this basis we learn how accurate our model based predictions are for new substances and data poor basins. The model train operates on the scale of Europe as a whole or for one or more individual river basins. It makes use of the pre-existing Europe-wide hydrology model HYPE. The Model Train complements lab and field based approaches, by providing information for substances and sites which are not included in monitoring and by providing full time coverage. Validation results for the Danube, Rhine, four Spanish and a series of Swedish River Basins reveal that the accuracy of the simulated concentrations of a range of chemicals is higher for substances with a single type of use (e.g. pharmaceuticals, pesticides) and lower for substances with multiple uses. The predicted chemical and ecological monitoring results, based on a correlation with the observed ecological status as EU Member States report it under the Water Framework Directive. The SOLUTIONS Model Train will offer an effective tool to screen a large number of chemicals on their impact on Europe’s aquatic ecosystems, and to do so with consideration for spatial and temporal gradients as governed by socio-economic and meteorological/hydrological patterns in combination with the chemicals’ physical and toxicological properties. The presentation will include the validation results and will highlight some of the Model Train application results from SOLUTIONS.

357 Forward-looking on possible impacts of chemical pollution: Modelling lethal and sublethal effects of chemical exposure on population viability for aquatic macroinvertebrates

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One challenge in the quality assessment of water bodies in Europe is the ongoing difficulty to link the chemical and the ecological status. Currently, new approaches are being tested to predict chemical and biological monitoring results, based on a correlation with the observed chemical impact on the ecological status of European water bodies in a retrospective way. Ecological modelling provides an alternative approach to connect exposure information to potential impact on biota, having the advantage that such modelling can be performed in a prospective way. This presentation will show results of ongoing modelling efforts in the EU 7th Framework Research Programme, which approaches are applied to link exposure dynamics of a number of chemical compounds to parsimonious individual-based population models. The STREAM-EU model provides exposure concentration results at the level of subcatchments, that is at a scale of tenths of km². Links between exposure and effects are realised using linear or log-logistic dose-response relationships. The ecological models account for lethal and sublethal effects on the population dynamics of a number of families of aquatic macroinvertebrates. Impacts of multiple chemicals are added up in the model following basic mixture modelling rules. Results depict the simulated inhibition of population growth rates and hence the chemical impact on population viability at European scales. Ecological modelling results are for some selected pairs of Europe compared with available monitoring information on the abundances of macroinvertebrate families in order to get an impression about the quality of the model predictions.

358 Eco-epidemiology of aquatic ecosystems: aligning chemical and ecological status

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In the framework of the COST Action 1301 the group has been working on the development and validation of a model to identify the ecological and health impact for emerging substances. The approach is validated for well-studied substances and data-rich basins. It makes use of the pre-existing Europe-wide hydrology model HYPE. The Model Train complements lab and field based approaches, by providing information for substances and sites which are not included in monitoring and by providing full time coverage. Validation results for the Danube, Rhine, four Spanish and a series of Swedish River Basins reveal that the accuracy of the simulated concentrations of a range of chemicals is higher for substances with a single type of use (e.g. pharmaceuticals, pesticides) and lower for substances with multiple uses. The predicted chemical and ecological monitoring results, based on a correlation with the observed ecological status as EU Member States report it under the Water Framework Directive. The SOLUTIONS Model Train will offer an effective tool to screen a large number of chemicals on their impact on Europe’s aquatic ecosystems, and to do so with consideration for spatial and temporal gradients as governed by socio-economic and meteorological/hydrological patterns in combination with the chemicals’ physical and toxicological properties. The presentation will include the validation results and will highlight some of the Model Train application results from SOLUTIONS.
Sustainability Environment and Health
This paper analyzes water quality assessment and management specifically looking at the diagnosis of toxic effects of mixtures in field ecosystems. We apply various methods to liaise ecosystem responses to mixture toxic pressure under natural, variable and multi-stressed conditions. We collated vast amounts of monitoring data to explore those diagnostically, in line with the Father of Epidemiology, dr. Snow, who famously provided a solution to a cholera outbreak in Soho, London, 1854, by epidemiological reasoning. So, this paper presents a set of contemporary eco-epidemiological results, the recognition of ecological impacts in surface water systems, and the diagnosis of probable causes. The paper presents the utility of that for chemical- and water quality assessment and management, thereby bridging preventive policies such as REACH via e.g. a Mixture Assessment Factor and environmental management issues such as IPPC Directive. We present novel results of scientific research at the nexus of chemical and water policies, connected to the European goals to reach a non-toxic environment and the good chemical and ecological status for aquatic systems. The presentation consists of the analyses of vast sets of surveillance monitoring data using a combination of techniques originating from the fields of bioassessment and ecotoxicology. It thereby bridges these—so far often disparate—scientific disciplines, to support sustainable chemical and water policies. One of the most recent examples is provided by a diagnostic analysis in which the Good Ecological Status appeared associated to the Good Chemical Status, the latter shown to be a limiting factor for reaching a good ecological status. The presentation will provide a rationale for eco-epidemiological analyses as well as various types of results, from diagnostics to prognostics to strategies. Further, this paper presents a body of evidence for the widespread occurrence of chemical mixture impacts in current water systems at the continental and national scale (Netherlands), corroborating recent findings of this kind based on chemical risk.

359 Unravelling the cocktail of stress: toxics and other stressors impacting on the ecological status of Europe’s rivers
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Water management requires solid understanding of how multiple stressors affect ecosystem status and services. The EU project MARS (Mixing Aquatic-conception and early stress characterization of chemical mixtures up to a solution-focused approach related to ecosystem services management goals. We present specifically a body of evidence for the wide-spread occurrence of chemical mixture impacts in current water systems at the continental and national scale (Netherlands), corroborating recent findings of this kind based on chemical risk.

360 Mitigation options for chemicals of emerging concern in surface waters: operationalizing solutions-focused risk assessment
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Chemicals of emerging concern (CECs) in the water cycle have been the focus of research for over a decade. They are currently added or used as stressors and adverse effects can be found in the environment. Future technological progress may help to identify future alternatives for the use and release of these chemicals, and therefore future developments are needed. This paper presents a specific approach to the mitigation of such criteria from the field of toxicology are available and well-corroborated. New and developing treatment technologies can be very promising but are generally not an option for end-users in water management as they need to have been tested on large scale and to be available commercially at relatively low cost. Commonly used advanced water treatment technologies are for sorption the use of activated carbon (granular activated carbon (GAC) and powdered activated carbon (PAC)), for (advanced) oxidation the use of ozone (O₃) and UV + H₂O₂ and finally the use of nano- and ultrafiltration membranes for size separation. We developed an evaluation criteria set for the specified treatment technologies. We used these criteria to evaluate removal efficiencies as collected in a database on removal efficiencies consisting of approximately 2000 entries, 93 compounds and 9 treatment technologies for wastewater (ozone, oxygen + H₂O₂, conventional WWTP, UV, UV + H₂O₂, PAC, GAC, NF, UF) and drinking water treatment (ozone, oxygen + H₂O₂, UF, UV + H₂O₂, PAC, GAC, NF, UF).

361 Future perspectives of chemical pollution and regulatory development
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Future developments in society will result in the emission of new substances to the environment which will require an adaptation of existing legislation for protection of human health and ecosystems. Scenarios for the future development of society can provide valuable indications on changes in future pollutants in river basins. Some developments are directly connected to consumption of specific substances, e.g. demographic change where a longer life expectancy will lead to changes in amounts and types of pharmaceuticals used and thus also to the related concentrations in the environment. Future technological progress may help to identify future alternatives for the use and release of these chemicals, and therefore future developments are needed. However, to prepare for a future with unknown scenarios for use and emissions of potential chemical pollutants is naturally difficult but a general approach can be developed and adopted to prepare for a future where environmental pollution by chemicals is avoided or minimised. This general approach should build upon a few basic principles: (1) The solution-focused approach: where the same approaches and evaluation options for minimising risks as to quantifying risks for new substances under development or introduction; (2) Transparency and openness of information and knowledge. Current applied research aimed at providing solutions to identified problems of chemical contamination in e.g. water ecosystems is severely limited by a lack of information on the production and use of chemicals in society as well as emissions to water. Linkage of national databases on use volumes of industrial chemicals such as SPIN (Substances in Preparations in Nordic Countries) would allow tracking quantitatively substitution of the most problematic substances; (3) Increased international cooperation and strengthened global agreements. The world is globalised and the transport of chemicals is transboundary — both via the atmosphere and via global trade.

Ecological risks under complex, multiple-stressor threat scenarios: integrating chemical effects with environmental drivers (I)

362 Towards a systematic approach for the assessment of multiple stressors: Making Aquatic Ecosystems Great Again (MAEGA)
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Climate Change Canada; K. Dafforn, Macquarie University / Evolution and Ecology Research Centre; L.E. Johnston, University of New South Wales / Evolution and Ecology Research Centre; K. Korbel, Macquarie University; D. Lappen, University of New South Wales; M. Mayer-Pinto, University of New South Wales / Evolution Ecology Research Centre School of Biological Earth and Environmental Sciences; W. Monk, Environment and Climate Change Canada; A. O'Brien, Wageningen University & Research; N. O'Brien, Environment and Climate Change Canada; K. Simon, R. Verdonschot, Wageningen University; P. van den Brink, Alterra/Wageningen UR / Aquatic Ecology and Water Quality Management Group

H. Barmentlo

In the Anthropocene, ecosystems are exposed to a range of stressors that if not properly managed can lead to ecosystem state shifts and significant losses in ecosystem services. We held a workshop (September 2017, Wageningen, The Netherlands) to develop a conceptual framework to assess the effects of multiple stressors on the structure and functioning of aquatic ecosystems. This framework was subsequently applied to three ecosystem types (ditches, floodplains and harbours). The proposed framework consists of two parts: an environmental filter and a transmitting function to allow effects to propagate to higher levels of biological organisation. Applying the framework consists of the following steps:

1) Select an ecosystem of concern;
2) Identify stressors and potential interactions;
3) Identify receptors/sensitive groups for each stressor;
4) Identify stressor-response relationships and group stressors according to their mode of action;
5) Construct an ecological model that includes relevant functional groups and endpoints;
6) Predict the resultant impact of multiple stressors;
7) Confront the predictions with experimental data;
8) Repeat until satisfactory.
different mode of action (the chemical pesticide Chlorpyrifos, CPF, and the biocidepesticide Bti in the mosquito Culex pipiens). We expected that the effects of the single exposures are strengthened in the presence of DTV. In addition, we tested whether there is an interaction between CPF and Bti and whether this interaction is magnified in the presence of DTV. We crossed three DTV treatments (no DTV, a small DTV of 7°C and a large DTV of 14°C) with four pesticide treatments (a solvent control, single CPF exposure, single Bti exposure and exposure to the CPF-Bti mixture). We investigated the effects on larval population growth rate ($r^*$) and its key components. The experiment was done in three steps: (i) 4-day exposure in L4 to DTV, (ii) 2-day exposure to DTV and the pesticide treatment and (iii) exposure to DTV until metamorphosis. The presence of a large DTV increased the toxicity (based on $r^*$) of the chemical pesticide, but not the biocidepesticide. Moreover, a large DTV changed the toxicity of the CPF-Bti mixture. For example, the presence of large DTV removed the antagonistic interaction effect on total mortality which was present in the absence of DTV and in the presence of small DTV. Our results underscore the importance of considering DTV as a factor shaping not only the toxicity of pesticides but also the interaction type between pesticides in mixtures. Given DTV occurs in all natural populations and may strongly differ between latitudes, DTV may be an important factor causing a mismatch between toxicity studies done in the lab at constant temperatures and the toxicity of pesticides and their mixture in the real world.

367 Warming and daily temperature fluctuations make the pesticide chlorpyrifos more toxic in Ischnura elegans damselflies J. Verheynen, R. van Roo, KU Leuven / Biology; R. Stoks, University of Leuven / Department of Biology
Current risk assessment of pesticides fails to protect aquatic biodiversity. A key reason is the lack of realism: pesticides are tested under ideal laboratory conditions at one mean temperature. To strengthen current risk assessment it is crucial to incorporate effects of global warming on the toxicity of pesticides. Global warming studies largely overlook that climate scenarios also predict stronger daily temperature fluctuations (DTFs), which can have greater fitness effects for organisms than increases in mean temperatures. While many pesticides (like organophosphates) get more toxic at higher temperatures, it is largely unknown how DTFs influence the pesticide toxicity. We examined a multiple-factor scenario where we quantified the single and combined effects of (i) increases in mean temperature and (ii) in DTF, and (iii) exposure to the pesticide chlorpyrifos (CPF) in larvae of high- and low-latitude populations of Ischnura elegans (CPF). CPF imposed mortality and more so in high-latitude compared to low-latitude larvae. Moreover, CPF was more toxic at 24°C compared to 20°C, confirming the higher toxicity of organophosphates at higher temperatures. A key finding was that DTF also increased the toxicity of CPF, providing novel evidence that DTFs can amplify the toxicity of pesticides. Furthermore, the increased toxicity of CPF by DTF was more pronounced at 24°C. This novel pattern is likely general as a higher mean temperature, DTF will expose the animals to even higher temperatures during the daily cycle, thereby increasing exposure to stressful temperatures. Also, the negative effect of CPF on larval growth strongly depended on DTF. CPF did decrease larval growth considerably, but only in the 10°C DTF treatment. Probably the higher metabolic demands for cell maintenance in the 10°C DTF treatment resulted in lower growth rates. Our results convincingly show that the organophosphate pesticide chlorpyrifos is not only more toxic to damselfly larvae at the higher mean temperature (24°C) but also at higher daily temperature fluctuations (DTFs) in terms of mortality and sublethal effects (growth rate). Notably, the synergistic effect of DTF on pesticide sensitivity was higher at the higher temperature. Our results highlight that incorporating higher mean temperatures and especially DTFs in ecotoxicology testing will increase the realism of the risk assessment of pesticides under global warming.

Trace organic compounds are important in environmental analysis, because they impact water quality and introduce potential (eco)toxicological effects. Current analytical methods mostly rely on gas chromatography (GC) or reversed-phase liquid chromatography (RPLC) coupled with (tandem) mass spectrometry. However, neither method can easily separate very small molecules. Two chromatographic separation strategies, a serial RPLC- hydrophilic interaction liquid chromatography (RPLC-HILIC) coupling and an analytical scale supercritical fluid chromatography (SFC) system will be presented, and their separation effectiveness as polarity-extended chromatographic methods for 274 environmentally relevant compounds were validated in a recent publication [1]. Compounds tested were grouped into three polarity classes, “very polar” log D (pH 7) below -2.5, “polar” log D (pH 7) between -2.5 to +2, and “non-polar” log D (pH 7) higher than +2. Nearly all compounds could be retained in both systems with relative standard deviations of retention times (RT) (n = 6) typically between 2% and 5%. Both techniques have considerable benefits when combined with accurate mass spectrometric detection. Molecules RT and accurate mass were recorded in a database for each set up. This information was used for compound screening measurements like “hidden-target screening” in complex environmental matrices (such as wastewater treatment plant effluents). SFC will complement and be useful for all types of molecules polarity. In this study, more than 80 percent of the compounds found in wastewater treatment plant effluent samples possessed a negative log D (pH 7) value. This result highlights the basic necessity to include “very polar” compounds in water monitoring techniques and protocols [1]. S. Bieber, G. Groen, S. Grosse, T. Letzel: RPLC-HILIC and SFC with mass spectrometry: Polarity-extended organic molecule screening in environmental (water) samples. Analytical Chemistry 2017, 89 (15), 7907-7914 (DOI: 10.1021/acs.analchem.7b00859).

369 Removal options and transformations of persistent mobile organic chemicals during production of drinking water A. TOUFFET, IC2MP CNRS; H. Gallard, IC2MP; B. Sieira, University of Santiago de Compostela; j. Chokki, b. tychene, IC2MP CNRS; R. Montes, University of Santiago de Compostela; R. Rodil, University of Santiago de Compostela; J. Quintana, University of Santiago de Compostela
Pesticides in mixtures. Given DTV occurs in all natural populations and may not be directly relevant to the natural environment, but DTV can be a threat to drinking water. Advanced treatments like activated carbon and oxidation processes can be used to limit the presence of organic micropollutants in drinking water. However, low removal by activated carbon is expected for PMOCs of their high polarity. The behavior of the plant effluent CPF and its degradation products were evaluated at lab-scale for their removal by different options including powdered activated carbon (PAC), high pressure membrane processes and transformation by ozone and chlorine. Highly polar PMOCs such as adamantane-1-amine (Log D = -2.34), trifluoromethanesulfonate (Log P = -1.35) and 1-caprolactam (Log P = 0.15) were not removed by PAC even for very high doses. Only naphthalenesulfonate (Log P = 0.41) was fully removed for 5 mg L$^{-1}$ PAC. The other PMOCs i.e. aromatic sulfonates, aromaticamines, phenols, were removed significantly for high PAC doses, but that are not compatible with drinking water production. Most of the PMOCs identified in water resources showed a very low reactivity with ozone with rate constants below 100 M$^{-1}$s$^{-1}$ and thus will not be transformed during ozonation of drinking water. Two aromatic guanidines, the 1,3-di-p-hydroxyguanidine and the 1,3-di-o-tolyguanidine, an olefinic sulfonate and an amine compound, the N-benzylendimethyamine, were rapidly transformed by ozone. Transformation of both guanidines occurred in few seconds at neutral pH during disinfection by chlorine. Chlorinated and hydroxylated analogues, and products of cleavage and cyclization were identified. US EPA toxicity prediction tool showed that chlorinated and hydroxylated analogues of PMOCs were less toxic than the parent compound, which was confirmed by Microtox acute toxicity test for Cl$_2$guanidine ratio of 1 and 10. Thus, reactions with chlorine during disinfection can be a source of new, persistent and more toxic chemicals in drinking water. Some PMOCs like 1-caprolactam, halogenated methanesulfonates, adamantane-1-amine and triazine compounds will neither be removed by adsorption on activated carbon nor by ozonation. Some PMOCs will be transformed by ozonation processes and could thus be present in drinking water. High pressure membrane processes would constitute the ultimate barrier for these compounds.

370 Removal of polar micropollutants from drinking water by reverse osmosis: a pilot scale study V. Albergamou, University of Amsterdam/IBED Institute / IBED; E. Cornelissen, KWR Watercycle Research Institute; B. Blankert, Oase; W. Knibbe, University of Wageningen; W. Van der Meer, Oase & University of Twente; P. de Voogt, University of Amsterdam / IBED
The occurrence of polar micropollutants (MPs) in drinking water sources is regarded as one of the most challenging issue of our times. Polar MPs can be used to limit the presence of organic micropollutants in drinking water. However, low removal by activated carbon is expected for PMOCs of their high polarity. The behavior of the plant effluent CPF and its degradation products were evaluated at lab-scale for their removal by different options including powdered activated carbon (PAC), high pressure membrane processes and transformation by ozone and chlorine. Highly polar PMOCs such as adamantane-1-amine (Log D = -2.34), trifluoromethanesulfonate (Log P = -1.35) and 1-caprolactam (Log P = 0.15) were not removed by PAC even for very high doses. Only naphthalenesulfonate (Log P = 0.41) was fully removed for 5 mg L$^{-1}$ PAC. The other PMOCs i.e. aromatic sulfonates, aromaticamines, phenols, were removed significantly for high PAC doses, but that are not compatible with drinking water production. Most of the PMOCs identified in water resources showed a very low reactivity with ozone with rate constants below 100 M$^{-1}$s$^{-1}$ and thus will not be transformed during ozonation of drinking water. Two aromatic guanidines, the 1,3-di-p-hydroxyguanidine and the 1,3-di-o-tolyguanidine, an olefinic sulfonate and an amine compound, the N-benzylendimethyamine, were rapidly transformed by ozone. Transformation of both guanidines occurred in few seconds at neutral pH during disinfection by chlorine. Chlorinated and hydroxylated analogues, and products of cleavage and cyclization were identified. US EPA toxicity prediction tool showed that chlorinated and hydroxylated analogues of PMOCs were less toxic than the parent compound, which was confirmed by Microtox acute toxicity test for Cl$_2$guanidine ratio of 1 and 10. Thus, reactions with chlorine during disinfection can be a source of new, persistent and more toxic chemicals in drinking water. Some PMOCs like 1-caprolactam, halogenated methanesulfonates, adamantane-1-amine and triazine compounds will neither be removed by adsorption on activated carbon nor by ozonation. Some PMOCs will be transformed by ozonation processes and could thus be present in drinking water. High pressure membrane processes would constitute the ultimate barrier for these compounds.
RO. Feed water samples were analysed by direct injection, whereas RO permeate samples where enriched by solid-phase extraction. The analysis were carried by ultrahigh-performance liquid chromatography coupled to time-of-flight high-resolution mass spectrometry. Neutral polar MPs displayed less than 5% passage, except benzoazolate, tollytriazole and phenylurea, which displayed a passage of 25%, 17% and 10%, respectively. The data showed that removal of neutral polar MP by RO is generally limited by size exclusion. For neutral and moderate polar MP the inverse was true as RO passage was well below the detection limit for bisphenol A, which displayed 4% passage. The higher passage of moderately polar and hydrophobic MPs could be attributed to solute-membrane hydrophobic interactions followed by diffusion. All anionic MPs displayed less than 1% passage, opposed to cations for which up to 10% passage was observed. The negative charge displayed by the membrane surface probably contributed to the results. Overall this study showed that high chemical removal rates can be achieved by RO. Tighter membranes and multi-stage RO will be investigated to improve the removal of small neutral MPs for drinking water applications.

371 Identification of transformation-derived very polar organic water contaminants and their relevance in the water cycle

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Highly polar organic substances are well water soluble, non-volatile, and exhibit only minimal adsorption to nonpolar surfaces. Therefore, they may be able to penetrate natural and artificial barriers and are thus mobile in the water cycle.1 If these mobile organic contaminants (MOCs) are persistent (POMCs) against microbial and chemical degradation, their removal during wastewater treatment and drinking water purification may prove difficult. Toxic POMCs can be classified as PMT (persistent, mobile, and toxic) substances. Since the most frequently used trace analytical method for the quantification of organic pollutants in aqueous matrices, reversed-phase high performance liquid chromatography - tandem mass spectrometry (RP-HPLC-MS/MS), is only of limited use for the analysis of very polar substances, little is known about MOCs in the water cycle and only few (e.g. acetaldehyde, glycolaldehyde) have been extensively studied and monitored2. PMOC may be, among others, industrial chemicals, or transformation products thereof. Most transformation processes usually result in the formation of transformation products (TPs) with increasing polarity until either mineralization is achieved or a dead end TP is formed, thus potentially resulting in persistent and highly polar water contaminants. Many PMOCs derived from transformation processes may still be unknown and thus not be represented in suspect or target screening campaigns. As a consequence, no information about their occurrence and origin is available, which severely exacerbates the sophisticated monitoring and effective regulation of their precursors. Based on the work of Arp et al.3 and Schueler et al.4 we selected 15 industrial chemicals with a high expected potential to further characterise the transformation and study their environmental behaviour during hydrolysis, biotransformation, oxidation with MnO2, and photolysis experiments. After structural elucidation of the 9 detected transformation products with high resolution mass spectrometry (HRMS) we developed a qualitative HILIC-MS/MS (Hydrophilic interaction liquid chromatography – MS/MS) method and screened 25 Hessian water samples for the presence of these TPs. While some TPs were not detected, others were observed in the majority of the major之事 the screening data provides first information about the potential environmental relevance of the identified TPs, which can be used to prioritize them for inclusion in future quantitative screening campaigns.

372 The limited chemical application domain of regulations: An illustration using the POP screening assessment in the Stockholm Convention

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Regulatory frameworks are initiated by a societal concern and built upon the chemical application domain, i.e. a chemical property space in which the frameworks can be applied in the context for which they were conceived and supported by a sound scientific foundation. As time passes, societal concerns change, and this can lead us to want to apply regulatory frameworks outside of their chemical application domain. Today we have the ambition to regulate tens of thousands of chemicals. Regulatory frameworks should therefore be designed in a way that they can be used to prioritize the management of chemicals based on their potential to impact the environment. However, regulatory frameworks are often limited by their existing scope and do not consider the potential impact of substances that are not currently regulated. This can lead to a misalignment between the intended purpose of the frameworks and their actual application. For example, the Stockholm Convention, which aims to eliminate the production and use of certain persistent organic pollutants, is often limited by its current scope and does not consider the potential impact of substances that are not currently regulated. Therefore, it is important to continuously reassess the relevance and effectiveness of regulatory frameworks and to adapt them as needed to address emerging chemical threats.

373 'One for all and all for one' - Can we REACH a harmonised PBT-assessment across EU-regulatory frameworks?

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Persistent, bioaccumulative and toxic (PBT) substances and REACH are frequently called in one breath. However, also other European regulatory frameworks for chemicals, such as for biocides, plant protection products (PPP), veterinary and human medicinal products (VMHP/HMP) stipulate the performance of a PBT-assessment during substance evaluation. As in other hazard based assessments (e.g. GHS/CLP regulation, POP), the PBT/PvP assessment focuses on the properties of a substance only and does not further take into account the use of the substance nor its exposure. Consequently, the identification of a PBT or PvP substance should be independent from the regulatory framework under which it is assessed. However, in our comparison of conclusions on PBT properties for a number of substances falling under more than one legislation it became apparent that the outcome of the PBT assessment does not necessarily correspond between different regulations. This stands in contrast to the goals to perform a comprehensible and consistent assessment of chemicals and to ensure a high level of protection of human health and the environment against hazardous chemicals within the EU. In order to elucidate the reasons still hampering a harmonised PBT-assessment, we did a compilation of a number of technical (amount and quality of data, acceptability of specific data, derivation of endpoints, applied guidance documents), and conceptual criteria (numerical criteria, testing strategy, assessment of transformation products) as well as of other factors (consequences of PBT-assessment, data management and publication of assessment results).

Outgoing from this, we developed several proposals facilitating a harmonised PBT assessment, starting from the implementation of an overall PBT-guidance up to an inclusion of transformation products in the PBT-assessment by all regulatory frameworks. Although it cannot be denied that a harmonisation process is ongoing, we conclude that there are still some fundamental choices to be made both at the organisational level and at policy level first to achieve the goal of a standardised PBT-identification among all legislations.

Product benefits and positive outcomes: valuation and beyond

374 A need for a better characterisation of product benefit in life cycle sustainability assessment

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Life cycle assessment (LCA) has become an important tool for assessing the environmental impact of products and production processes. It allows for a comprehensive evaluation of the environmental footprints of goods and services, from raw material extraction to production and disposal. However, there is a need for a better characterisation of product benefit, which is a key aspect of LCA. Product benefit refers to the positive impact that a product has on the environment, society, and economy. It is important to consider product benefit in LCA because it allows for a more comprehensive assessment of the overall impact of a product across its entire life cycle.

In consequential LCA, not only the consequences related to the use of the product but also those related to its production are considered. This means that the entire life cycle of a product, from raw materials extraction to disposal, is taken into account. This approach provides a more holistic view of the environmental impact of a product and allows for better comparisons between different options. However, current LCA frameworks often focus on the consequences of using a product, rather than considering the consequences of producing it. This is a limitation because the production of a product can have significant environmental impacts, which may be overlooked if only the use phase is considered.

A need for a better characterisation of product benefit in life cycle sustainability assessment

Third, when an additional amount of byproduct is created by a production system, this may also induce a byproduct benefit, which can be
accounted for as such, in fact, when a byproduct enters the market, a share of it can lead to a decrease in supply (substitution approach) but another share can also lead to an increased demand and thus consumption, which satisfies needs that were previously unsatisfied (production benefit approach). A consideration of both effects is needed in CLCA.

375 Assessing nutritional impacts and benefits on human health in LCA: A new midpoint impact category
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Decision of a crucial determinant of human health. According to the Global Burden of Disease (GBD), dietary risk factors are responsible for >10 million deaths/year globally. Yet, beneficial and detrimental nutritional health effects, the dominant pathway for health in food systems and diets, are often neglected in Life Cycle Assessment (LCA). To address this, we develop 14 nutritional characterization factors (CFs) for food groups and nutrients using epidemiological evidence from the GBD, and propose a new nutritional midpoint impact category for LCA, demonstrating its application to the entire US diet. We develop marginal nutritional CFs for 8 major food groups (nuts and seeds, whole grains, fruits, vegetables, milk, sugar-sweetened beverages, red meat, and processed meat) and 6 nutrients (omega-3, calcium, fiber, polyunsaturated fats, trans fats, and sodium), identified by the GBD as dietary risk factors. CFs are estimated by coupling age- and gender-adjusted information on outcome-specific incidence rates with risk ratios (RR) and severity factors, measuring positive or detrimental effects in avoided DALY/y. We also develop a profiling system for 6000+ food items consumed in the US that allows publically available data from multiple databases with risk factor definitions from the GBD. Finally, for 6000+ food items we estimate the HEalth Nutritional Index (HENI), the total avoided health burden from all dietary risk factors per serving and 100 kcal. Nutritional CFs for food groups and nutrient range between -8 avoided DALY/y for sodium, up to 57 avoided DALY/y for omega-3 from seafood. HENI score typically ranges from -80 avoided DALY/serving for Frankfurter sandwiches to 50 avoided DALY/serving of nuts and seeds. Absolute HENI scores and ranking of food items vary substantially when using 1 serving or 100 kcal as a functional unit. Unhealthy food groups such as mixed dishes and protein foods with the exception of seafood and nuts and seeds have negative HENI scores primarily due to detrimental effects from processed and red meat, saturated and trans fats, and sodium. Healthy food groups such as fruits, grains, and vegetables excluding starchy vegetables tend to have positive HENI scores dominated by health benefits from their respective food groups. The estimated nutritional CFs for a new midpoint impact category in LCA would improve human health impact assessment in LCA and allow for a comprehensive assessment of food items and diets.

376 Combining Operational Research and Life Cycle Assessment to optimize the environmental performance of Peruvian diets
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Food production and security has been highlighted as one of the most threatened sectors worldwide due to damages of climate change. However, food production is also responsible for an important fraction of GHG emissions. In Peru, 50% of household expenditure is destined to food purchase. In contrast, malnourishment is still rampant in many socioeconomic sectors, mainly in the Highlands in the Amazon basin. In this context, it appears as a major challenge to jointly achieve nutritional improvements in the Peruvian diet and reductions in terms of GHG emissions. Hence, the main objective of this study was to apply a methodology which allowed optimizing the environmental profile of the Peruvian diet while improving its nutritional requirements at competitive economic costs. In other words, the aim of the optimization model was to determine an optimal diet from an environmental perspective considering nutritional and economic constraints. For this joint combination of Operational Research and Life Cycle Assessment was performed. Based on the average diets identified for each city included in the study, an optimization was performed considering a set of criteria that respond to the three dimensions of sustainability. Nutritional aspects were included in the model through a restriction based on the minimum consumption of food types and caloric intake recommended by Peruvian authorities. Regarding economic costs, the model included a set of inequations that limited the minimum and maximum monetary changes throughout the year (i.e., 2016). Finally, environmental aspects were considered by introducing an objective function that minimizes the emissions of CO₂eq of the entire food basket. The result of the proposed linear program allows understanding the amount of each individual food product that should be consumed in each city that satisfy all the restrictions included in the model in order to attain the lowest GHG emissions possible. AMPL® was used as the programming platform, and CPLEX® as the solver. Results demonstrated that substantial reductions can be attained in GHG emissions through the optimization of diets in Peru. For instance, in Lima the reduction could reach 200 kg CO₂eq per person and year (22%). These results constitute an important framework to understand the current situation of the GHG emissions of the average Peruvian diet, as well as to mitigate these emissions while improving nutritional aspects and controlling economic costs.

377 Using the first Swiss dietary survey to determine the environmental and health benefits and impacts of various dietary patterns
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Many studies compare the environmental impacts of dietary patterns such as dietary recommendations (e.g. food pyramids), vegetarian and vegan diets. Mounting evidence suggests diets high in red meat and other animal products have higher associated environmental impacts. A hypothetical non-meat diet is often considered in such assessments, which may e.g. be equi-calorie or mass to the meat containing diet. In this study we use the first Swiss National Survey (MenuCH) to determine what non-meat eaters consume in Switzerland and what potential environmental and health benefits (or impacts) may result from assessing realistic consumption. About 5% of the Swiss population self-identifies as vegetarian, and less than 1% as vegan. Meatless diets contained about the same overall mass of food consumed, generally offered environmental and health benefits through increased fruit and vegetable consumption, but vegan diets can be insufficient in certain essential vitamins if not supplemented. Nuts, seeds, and their oils were important sources of key nutrients such as vitamin E. In conclusion, using dietary surveys can help provide us with evidence as to what people are eating. Our study looks into the monetary and other animal products from the diet. Accounting for the environmental and health benefits of realistic dietary patterns, can help support improved recommendations.

378 The cost of CO2 in Life Cycle Assessment
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Climate change has gained increasing attention over the past decade in response to the revelation that we need to maintain a viable climate for humans and the environment. The increasing emission of greenhouse gases (GHG) such as CO₂ may accelerate climate change and cause subsequent damages. Correspondingly, countries and companies actively develop strategies to minimize their GHG emissions and thus climate impacts, but which strategies that will be more beneficial is often hard to evaluate. Life Cycle Assessment (LCA) is a tool to evaluate the damages of GHG emissions from the whole life cycle of the intended strategy, taking a cradle-to-grave perspective. By monetising the impacts related to these emissions, they can be compared to the overall cost of a strategy. This secure that emissions are considered in determining the priority and benefits of the assessed reduction option. Our study looks into the monetary values of GHG, represented by CO₂ or CO₂-equivalent, and their underlying cause-effect chains in three Life Cycle Impact Assessment (LCIA) methods LIME2, EP2015 and ReCiPe2016. The damage cost for CO₂ is in the same order of magnitude in EPS2015 and ReCiPe2016, but one order of magnitude higher than that in LIME2. Climate change-related damages on human health are well represented in all LCIA methods, and the monetised damages from this category contribute to more than 70% of the total CO₂ cost in all three methods. Social assets and ecosystem damages, on the other hand, are only counted for in two of them. Furthermore, a range of potential socio-economic damages from a changing climate are discussed in IPCC reports, including economic loss from extreme weather events, costs of potential climate-related society security and poverty, but they are not included in any of the LCIA methods. This may limited the suitable application area of the CO₂ cost evaluated by LCA, especially in studies where social and economic consequences are the major concerns. The CO₂ costs from the three LCIA methods are further evaluated in comparison with approaches from other research fields, such as Social Cost of Carbon (SCC), and discrepancies and associated uncertainties are discussed.

379 Poster spotlight: WE257, WE258, WE259

Advances in monitoring and evaluating remedy effectiveness for in situ amendments in soils and sediments

380 The assessment of Human Health Benefits and Risks of Contaminated Sediment Remediation
J. Kvasnicka, University of Michigan, Ann Arbor USA / Environmental Health Sciences; K. Stylianou, University of Michigan - School of Public Health /
Six inches under: Remediation efficiency of activated carbon caps buried by dynamic sediment movement

S. Abel, J. Akkanen. University of Eastern Finland / Department of environmental and biological sciences

The placement of activated carbon (AC) based thin layer caps is a promising and novel in-situ remediation method for contaminated sediments. The method utilizes the adsorptive capabilities of AC, allowing it to strongly bind persistent organic pollutants, thus greatly reducing the contaminants’ bioavailability and sediment to water flux. Sediment capping with active materials can furthermore provide a less destructive, low-cost alternative to traditional remediation methods, such as sediment dredging. In this study, the method was rigorously evaluated, later realistic, conditions with the first field trial on AC based sediment remediation in Finland. At the test site in the PCB-contaminated Lake Kernaalanjärvi, a 300 m² plot was amended with an AC thin layer cap (1.6 kgAC/m²). Due to the shallow nature of the lake and its large surface area, highly dynamic sediment movements occurred over the monitoring period of 14 months. As a consequence, the AC cap was buried rapidly under a layer of transported sediment, leading to a low measurable impact of the AC amendment. Neither bentic community structure nor PCB bioaccumulation in local benthic organisms were significantly different on the AC amended plot and the surrounding reference sites. The amount of sediment deposited was high (34.2 g dry matter/m²/day), making detailed observations on the development of the situation over time difficult. To investigate the potential for a long term remediation success of AC caps even under these unfavorable conditions, a corresponding laboratory study was conducted. The field scenario was replicated in test vessels containing a base layer of PCB contaminated sediment topped with an AC layer that was in turn buried by varying amounts of sediment (thicknesses of the overlying sediment layer ranged from 1 to 40 cm). The endpoints were the growth and PCB bioaccumulation in Lumbricus variegatus and Chironomus riparius. The results indicate that an AC cap can remain effective even once it has been covered with contaminated sediment. This depends, however, on the intensity and depth of bioturbation. With the deeper dwelling L. variegatus present in the test systems, the AC layer was mixed well with the overlying sediment, allowing for a measurable remediation success. With the shallow dwelling C. riparius, this effect could only be observed with minimal sediment coverage of the AC (<5 mm).

Ecosafe nanotechnologies for environmental remediation: the NANOBOND project

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In situ remediation of sludge and dredged harbour sediments is currently highly cost-effective despite an ever increasing number of sites requiring swift treatments to reduce contamination. Therefore there is a urgent need worldwide to identify new promising and innovative solutions ensuring a quick and efficient removal of pollutants and appropriate methods for monitoring the effectiveness of remediation strategies and of uncertainties associated with these potential adverse effects are usually not quantified in a manner conducive to comparing them to potential benefits and assessing overall remeedy effectiveness. Focusing on the Hudson River PCBs Superfund Site remediation, this study demonstrates a novel approach to comprehensively evaluate the relative tradeoffs between population health benefits and risks associated with different remedial alternatives. The specific aims were to: 1) Assess health impacts on recreational anglers for a No Action scenario, due to bioaccumulation of PCBs in Hudson River fish, and exposure through fish consumption. 2) Determine and compare the reduction in health impact from reduced fish tissue PCB concentrations associated with different remedial options relative to No Action. 3) Investigate potential health impacts of the selected remedy from resuspension of sediment, air emissions of PCBs, diesel particulate matter (DPM) and NOx, and occupational accidents. 4) Compare the avoided health impacts, i.e., the health benefits, with the created health impacts. For each considered impact pathway, we derived both central and upper bound estimates, using the disability adjusted life year (DALY) as a comparative metric. For the No-Action scenario, the health impacts are 11 and 78 DALY’s, respectively. The power of this study is its potential to be extended to other contaminants and sites, with the potential to generate more detailed observations on the development of the situation over time difficult. To this end, the NANOBOND (Nanomaterials for Remediation of Environmental Matrices - Environmetal Assessment - Remediation of matrixes - Environmental nanotechnologies for environmental remediation, known as nanoremediation, represents a challenging and innovative solution, environmental and human risk assessment associated with the use of ENMP are still a matter of debate. Limited in situ applications to water and soil remediation are due to lack of data regarding environmental impacts and there is a general demand for strategies aimed to fulfill such needs. As the potential and efficacy of nanotechnologies is well established, several drawbacks related to full-scale application should be overcome. In particular, great efforts should be devoted to develop (nano)materials which own eco-friendly features such as limited release and mobility in environmental matrices as well as no toxicity for natural ecosystems. Eco-effective can be thus used to develop ecofriendly (nano)materials for environmental application and to provide monitoring methods in a weight-of-evidence approach also to support decision-makers. The NANOBOND project (Nanomaterials for Remediation of Environmental Matrices associated to Dewatering) funded in 2015 by POR CRoF FESR Toscana 2014-2020, is developing an innovative system for treating contaminated sludge and contaminated water by using AC caps to quickly and cost-effectively remove contaminants. The method utilizes the adsorptive capabilities of AC, allowing it to strongly bind persistent organic pollutants, thus greatly reducing the contaminants’ bioavailability and sediment to water flux. Sediment capping with active materials can furthermore provide a less destructive, low-cost alternative to traditional remediation methods, such as sediment dredging. In this study, the method was rigorously evaluated, later realistic, conditions with the first field trial on AC based sediment remediation in Finland. At the test site in the PCB-contaminated Lake Kernaalanjärvi, a 300 m² plot was amended with an AC thin layer cap (1.6 kgAC/m²). Due to the shallow nature of the lake and its large surface area, highly dynamic sediment movements occurred over the monitoring period of 14 months. As a consequence, the AC cap was buried rapidly under a layer of transported sediment, leading to a low measurable impact of the AC amendment. Neither bentic community structure nor PCB bioaccumulation in local benthic organisms were significantly different on the AC amended plot and the surrounding reference sites. 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With the deeper dwelling L. variegatus present in the test systems, the AC layer was mixed well with the overlying sediment, allowing for a measurable remediation success. With the shallow dwelling C. riparius, this effect could only be observed with minimal sediment coverage of the AC (<5 mm).
been detected at fairly high levels in aquatic systems (0.33-611 ng/L), terrestrial environments (0.53-340 µg/kg), and in the tissue of organisms (4.6-23.6 µg/kg in crop tissues, 61-127 µg/kg in terrestrial invertebrates) (Chen and Ying, 2015; Kinney et al., 2006; Pan et al., 2014). Long-term exposure to the residues of polychlorinated biphenyls could pose a risk to the ecological system and exert adverse effects on human health via food chain (Carvalho et al., 2014). Adsorption processes have a decisive role for the environmental behavior and the ultimate fate of pharmaceuticals (Drilla and Lyberatos, 2005). However, relatively a few investigations of the sorption of organic compounds at the group level based on the dissociation degree of molecule in soil have been published so far (Droge and Goss, 2013; Franco and Trupp, 2008; Franco et al., 2009; Kah and Brown, 2007). The main aim of this study was to explore the effects of properties of the chemical and soil characteristics on the sorption of some organic compounds in the soil environment. First, sorption behaviors of nineteen pharmaceuticals across four groups ( neutrals, strong bases, weak bases, acids) were explored in five test soils. Using the measured sorption coefficients for each group, we evaluated the applicability and accuracy of existing predictive models that have been proposed to predict the sorption behavior of organic chemicals in soil. Finally, Pearson correlation analysis and Principal components analysis (PCA) have been carried out at the group level to systematically assess the potential factors (both soil and drug properties) influencing the sorption behavior of pharmaceuticals in soil and to get better understandings of the sorption mechanisms of different pharmaceuticals in the soil.

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In vitro and in vivo assays to evaluate chlordecone transfer to animals: interest of soil amendment

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Chlordecone (Kepone) (CLD) is a highly persistent pesticide formerly used in French West Indies. Nowadays high levels of this pesticide are still found in soils which represent a subsequent source of contamination for outdoor-reared animals. In that context, sequestering matrices as activated carbons (ACs) or biochars are believed to efficiently decrease CLD transfer to animals. The present study intends to test using 2 distinct in vitro tests prior an in vivo assay the respective efficiency of several biochars and ACs to limit CLD transfer to animals. The Te-PBET and the ISO/DIS 16 751 availability part A protocols were used. In each test amended soils were prepared from a control one (SS) by adding 2% (mass basis) of one of the ACs or biochar. A selection of interesting matrices was realized prior the in vivo part of the study. Four animals were used for each of the contaminated soils. Only treatment groups exposed through amended soil with ACs presented significant decreases CLD availability, bioaccessibility (< 8%). Similar results were found using both in vitro assays. At last, concentrations of CLD in piglets liver and adipose tissue were found significantly lower after exposition to an AC amended soil (p < 0.001). This decrease was particularly high for a coconut shell activated carbon where relative bioavailability was found lower than 3% for both tissues. Finally, a positive correlation was found between environmental availability, bioaccessibility tests and in vivo results. This study leads to conclude that (i) AC introduced in CLD contaminated soil should strongly reduce CLD availability; bioaccessibility and bioavailability (ii) Tested biochars showed no reduction of transfer (iii) availability and bioaccessibility tests could be useful screening tests in order to select the appropriate biochar or AC.

Analysis and Fate of Emerging Contaminants in soils, water and plants under water scarcity (I)

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Determination of dioxin-like polychlorinated biphenyls in land near the dumps of some settlements of the Republic of Armenia

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The sources of environmental pollution with polychlorinated biphenyls along with the energy production/ distribution complex include landfills, many of which do not correspond to environmental requirements and are the only attribute of the “consumer” relationship to nature, thus bringing forth a number of problems and a great danger for the normal functioning of biocenosis. In the present research we used samples of the following pollutants from the landfills and agricultural fields or water basins near some settlements of the Armenia. The following 14 dioxin-like polychlorinated biphenyls (PCBs) were determined in soil samples: congeners NN 77, 81, 105, 114, 118, 123, 126, 156, 157, 167, 169, 170, 180, 189. Quantitative determination was carried out using chromatography with electron capture detectors (ECD) equipped with glass capillary column with stable phase DB-5MS UI and the following parameters: 60 m x 0.250 mm x 0.25 µm. Special attention was paid to the total amounts of polychlorinated biphenyls, as the total amount of these compounds correlate with the hygienic standards, which as such are integral values. In all investigated soil samples dioxin-like PCBs were detected, however, in this case we mainly recorded congeners NN 77, 81, 105, 114, 118, and 123, while NN 169 was determined less frequent and at insignificant quantities. Attention was drawn to the following: - out of 7 randomly selected soil sampling sites, a 2 to 3.5 times exceeding of the total/summary standard level was found at four sites; - in all cases, the excess of the standard was due to PCBs NN 81 and 114. Of special attention is the fact that all of the soil sampling sites (Dilijan Town, Tavush Province of Armenia) along with 3.5-fold exceeding the standard, almost all dioxin-like PCBs were found.

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Associated Health Effects of Veterinary Pharmaceutical Residues in Wastewaters around Selected Livestock Agriculture Farms in Western Cape Province

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Livestock farming is a major industry in the Western Cape Province of South Africa and livestock agricultural farms have been suggested to be a major source of pharmaceutical residues in many aqueous matrices, due to their enormous use. Pharmaceutical products such as steroids and non-steroids hormones, antibiotics and non-steroidal anti-inflammatory drugs from agriculture have been indicated to have the potential to show significant endocrine and other health effects. In this study, a High Performance Liquid Chromatography coupled to ultraviolet detector (HPLC-UV-Vis) method was optimized and validated for the detection and separation of the selected pharmaceuticals in effluents for livestock farms with major activity in the study areas. Multi-residue solid phase extraction (SPE) procedure was developed and validated for the recoveries of acetaminophen (AC), diclophenac(DP), salicylic acid (SA), tetracycline(TC), chloramphenicol(CHR), ciprofloxacin(CP), bisphenol-A (BPA), estradiol(II), oestrone(IV), oestradiol(V), progesterone(PBET) and the respective efficiency was assessed in effluents from livestock farms. The results of the health risk assessment clearly showed mutagenic activity being observed in samples from sheep and poultry farms. It also showed higher aglycogenic activity in the pig farm. The results indicated that making use of the maximum concentration of 17β Estradiol found in the samples, there was a slight risk of developing cancer through accidental ingestion via recreational activities with higher risk if the water was used for domestic purposes without treatment to remove them or if the water was used for irrigation purposes.

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Characterization of respective contribution of agricultural and urban sources to pesticide contamination of a peri-urban river

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Water is nowadays a precious resource on which anthropic pressure increased drastically these last years, due to global lifestyle improvement and the population growth. Pesticides are part of the most preoccupant micropollutants in aquatic environments because of their intrinsic toxicity, even at trace-levels. They were firstly used for agricultural yield improvement but they are now used as biocides for the protection of construction materials, wood, textiles, paints, etc., or as veterinary treatment susceptible, and can be discharged in rivers via wastewater treatment plants (WWTP) or Separated Stormwater Overflow (SSO). This multiplicity of uses is linked to high concentrations in rivers, affecting aquatic ecosystems that play role of final receptacle for micropollutants in general. It is thus necessary to consider pesticide impact to water ecosystems. Treatment of pesticides can be quite expensive and inputs may not be clearly identified or collected. On the reduction at source can be considered as an interesting alternative (dose reduction, practice changes, etc.). However this approach requires first of all the identification of uses responsible for inputs, as few information is available nowadays. This study monitored for 4 years a peri-urban continuum formed by a river, SSO and a WWTP also as wastewaters to link uses and presence in environment. Water bodies presented distinct contaminations profiles: rivers were characterized by important concentration of plant protection products while WWTP effluents presented important concentrations of biocides and veterinary molecules which are among the most toxic pesticides. Flux calculation allows identifying agriculture as the major source of plant protection products while WWTP brought the most part of biocides and veterinary products, especially in low-flow period when the WWTP contributes up to 40% to the overall flow of the studied river. Storm sewers had an intermediate status, with less consequents inputs but are still significant because of lack of treatment on these effluents and a potential increase of concentration around the
Phragmites australis enantioselectively uptake, translocate and degrade the chiral pesticides tebuconazole and imazalil.

Environmental risk assessment approaches often require information on the free concentration in water, bioaccumulation factors in aquatic organisms, by-products and their toxicity in order to determine the effect of a contaminant on ecosystems. Thus, in the present work, in order to assess the uptake, distribution in different tissues (liver, muscle and gill) and bio-fluids (plasma and bile), metabolism and elimination of BP-3 in gill-head bream (Sparus aurata), a controlled dosing 14-day experiment was designed at 50 ng/mL concentration level. BP-3 was detected in all the analysed samples, with the highest concentrations at day 14. Bile concentrations were significantly higher in comparison to the rest of tissues/fluids. Since BP-3 is hydrophobic and non-ionizable compound, the lowest concentrations of BP-3 were found in plasma. Although liver tissue (highly lipidic) could be an appropriate reservoir of BP-3, the low concentration of non-metabolized BP-3 found in this tissue could indicate a high metabolism activity in liver. And on the contrary, the lack of biodegradation activity in muscle (less lipidic) can explain the second highest concentrations detected, reaching the equilibrium state in the 4th exposure day. In any case, the occurrence of BP-3 in gills suggests that at least part of the uptake occurred through the gills. To completely characterize BP-3 exposure, the analysis performed by means of liquid chromatography – high-resolution mass spectrometry allowed the identification of a broad suite of BP-3-by-products in serum, water and fish tissue/biofluids (mainly in bile and liver). By the interpretation of the MS2 spectra, we identified demethylation, hydroxylation and glucuronidation as the main degradation pathways of BP-3. Acknowledgements - This work was financially supported by the Ministry of Economy and Competitiveness through the projects CTM2014-56628-C3-1-R and CTM2014-56628-C3-2-R, Xunta de Galicia (ED431C201373/36) and FEDER/ERDF. H. Ziarrusta is grateful to the Spanish Ministry of Science and L. Mijangos to the Basque Government for their predoctoral fellowships.
393 Estimation and prioritization of hospital API emissions
A. M. Ragas, Radboud University / Department of Environmental Science; C. van Lenteren, M. Galpen, K. Tipatet, Radboud University; R. Oldenkamp, Radboud University / Department of Environmental Science

Hospitals constitute an important source of APIs, particularly for substances like antineoplastics and contrast agents. Measuring these emissions and their impacts is possible, but is very time-consuming and costly. The main aim of the present study was to develop an approach for estimating hospital API emissions based on hospital purchase data and to prioritize these emissions based on potential environmental impact. A model was developed to estimate the API loads reaching the hospital sewer system. The model accounts for the return of unused APIs, route-specific excretion by patients, non-patient API use (e.g., personnel) and off-side emissions. The model was operationalized for 16 APIs emitted by two academic hospitals in the Netherlands. Model predictions were validated based on measurements of APIs in the sewer system using passive samplers with speediaks as absorbent. The samplers were deployed over a 10-12 day period and analyzed in the laboratory using LCMS. Most of the estimated loads were within a factor of 10 of the measured loads. On average, estimations for Hospital 2 were more accurate than for Hospital 1, which was probably due to the use of monthly purchase data and some other small model improvements implemented for Hospital 2. APIs which are typical for hospitals (e.g., antineoplastics and contrast media) were relatively well predicted. The prioritization of the APIs based on environmental impact was substantially influenced by the availability and interpretation of toxicological data. Diclofenac ranked highest, but this ranking was determined by one particular toxicity study of which the validity is being disputed. Ciprofloxacin consistently ranked high, and to a lesser extent also paracetamol and metoprolol. Azithromycin and imipenem also ranked relatively high, but only limited toxicity data were available for these substances, resulting in large safety margins.

394 Development and validation of a model to predict concentrations of human APIs in European surface waters
R. Oldenkamp, S. Hoeks, V. Barbarossa, M. Cengic, Radboud University / Department of Environmental Science; L. Carter, University of York / Environment Department; E.E. Burns, University of York / Chemistry; J. Wilkinson, The University of York / Natural and Built Environments; A. Boxall, University of York / Environment Department; A. M. Ragas, Radboud University / Department of Environmental Science

Active Pharmaceutical Ingredients (APIs) are consumed in large quantities, and end up in the environment as a result of not being degraded completely during passage through the human body and wastewater treatment plants (WWTPs). Although reported concentrations are generally low, adverse ecological effects caused by some human APIs are plausible considering their specific modes of action and high potency. Consequently, the issue of human APIs in the environment has been acknowledged as an emerging environmental problem requiring scientific and regulatory attention. A crucial step in environmental risk assessment of APIs is the estimation of their environmental exposure potential. Since there are currently more than 4,000 different human APIs in use, monitoring individual APIs is practically impossible. The aim of the present study was to develop and validate a model for the prediction of environmental concentrations of APIs from country-specific data and characteristics (e.g., antineoplastics and contrast media) were relatively well predicted. The prioritization of the APIs based on environmental impact was substantially influenced by the availability and interpretation of toxicological data. Diclofenac ranked highest, but this ranking was determined by one particular toxicity study of which the validity is being disputed. Ciprofloxacin consistently ranked high, and to a lesser extent also paracetamol and metoprolol. Azithromycin and imipenem also ranked relatively high, but only limited toxicity data were available for these substances, resulting in large safety margins.

395 Occurrence and fate of the antidepressant metformin and its transformation products
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Effluents of municipal wastewater treatment are major entry pathways for pharmaceuticals and their transformation products (TPs). A wide-spread compound is the antidepressant drug metformin (MF) with its well-known main metabolite guanyl urea (GU). GU is formed in WWTPs. So far, no other TPs of metformin are reported in the water cycle. In this study, electrochemical experiments for simulation and identification of potential new TPs of MF were performed. In addition we investigated the occurrence and fate of MF and its TPs in WWT and surface water. Analysis was performed by LC-high-resolution-mass-spectrometry (HRMS) using HILIC (hydrophilic interaction chromatography) quadrupole-time-of-flight mass spectrometry (QTOF-MS). Four TPs of MF have been identified after electrochemical degradation. The proposed structures are 4-amino-2-imino-1-methyl-1,2-dihydro-1,3,5-triazine (4,2-AIMT), 2-amino-4-methylamino-1,3,5-triazine (2,4-AMT), 2-diamo-1,3,5-triazine (2,4-DAT) and methylbguanidine (MBG). The mass error was below 3 ppm for all 4 TPs. However, the well-known TP GU could not be formed electrochemically. The TPs found are similar to those of a former study using gamma radioisotopes (Collin et al. 2004). 24-hour mixed samples of wastewater in Southwest Germany were obtained for 7 consecutive days. Elimination of MF was 92 â¢ at an average influent concentration of 24 µg/l. GU concentrations were in the influences between 66 and 640 µg/l and in the effluents between 60 and 386 µg/l. A plausible reason for the occurrence and relatively high concentrations of GU compared to MF could be the formation of GU already in parts of the sewer system. The following oxidation products of MF have been detected for the first time: 2,4-DAT, MBG and 2,4-AMT. The concentrations of MBG ranged between 40 and 122 ng/l. For the other TPs no authentic standards were available, however TPs 2,4-DAT and 2,4-AMT showed similar mass spectral fragmentation patterns, which implies their formation during WWT. 3 grab samples of surface water affected by waste water showed relatively high MF (between 100 and 470 ng/l) and GU (between 3700 and 4500 ng/l) concentrations. MBG was in the range between 10 and 30 ng/l. In addition, 2,4-DAT was detected. Its response was in all three samples about 40 % of the response of the WWTP effluent samples. The study was performed within the project “Effect-Net”, funded by the Ministry for Science, Research and Art, Baden-Württemberg.
The University of York / Natural and Built Environments; R. Ashauer, University of York / Environment; A. Boxall, University of York / Environment Department

The majority of active pharmaceutical components (APIs) currently in use are ionisable and may become charged at environmentally relevant pHs. Previous research has shown that the accumulation of a molecule in aquatic invertebrates depends on the ionisation state of the molecule which is driven by the pH of the surrounding medium. Recently a toxicokinetic modelling approach has been proposed to account for accumulation rate. Assessed were metabolic and phytototoxicological effects. The number of flowers and plant height were assessed for selected species to evaluate differences in development and flowering. In addition, seeds were sampled to evaluate differences in seed quantity and quality. Furthermore, the results will be compared to a non-target terrestrial plants pilot field study (Knaebe et al. 2017; Presentation at SETAC Europe 2017). References: EFSA PPR Panel (2014). Scientific Opinion addressing the state of the science on risk assessment of plant protection products for non-target terrestrial plants. EFSA Journal 2014;12(7):3800, 163 pp. OECD (2006). Test No. 208: Terrestrial Plant Test: Seedling Emergence and Seedling Growth Test. OECD Publishing, Paris. OECD (2006). Test No. 227: Terrestrial Plant Test: Vegetative Vignour Test. OECD Publishing, Paris.

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Predicting plant community level effects of herbicides based on monoculture dose-responses: Testing the plant community model IBC-grass with experimental data

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Ecological models are rarely found in terrestrial plant ecotoxicology and risk assessment. Especially on community-level, the number of suitable plant models is scarce. Existing models are often not validated with experimental data, although the validation of ecological models is an important point for their credibility for risk assessors. Nevertheless, ecological models are a suitable tool to extrapolate individual-level greenhouse experiments to the plant communities in the off-field. Especially bearing in mind that protection goals for non-target terrestrial plants as defined by EFSAs are on population and community level. Reuter and Siemoniet-Gast (2007) performed an experiment that includes not only the test of monocultures but also the test of small artificial communities consisting of 6 different plant species. This study is suitable to be coupled and compared to a plant community model, since it covers the population as well as the community level. We adapted the plant community model IBC-grass to the settings of the empirical study by Reuter and Siemoniet-Gast (2007). Specifically, we analyzed to which degree the model is able to represent realistic community level effects not only by comparing visual patterns but also by calculating model adequacy and reliability as a measure for the model fit. The predicted effects of the model IBC-grass show a good agreement with the experimental data, for the monocultures as well as for the communities. Model adequacy was lower in the monocultures. However, model adequacy increases in the communities. In general, model reliability is high in the monocultures but low in the community setup. This may be due to the test of a non-standard species. Most tests were performed based on the OECD guideline OECD 221, the two Myriophyllum spicatum guidelines OECD 238 and OECD 239, the ASTM E1913-04(2012) for Myriophyllum sibiricum, Sediment contact test with Myriophyllum aquaticum (ISO/DIS 16191) and the principles of the method proposed by the AMRAP (Aquatich Macrophyte Risk Assessment) guideline. Although the model can serve as an important tool for predicting potential impacts on natural environments with information obtained from greenhouse studies on individual species.

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Use in risk assessment of recovery in plants from exposure to chemicals

T.A. Springer, EAG Laboratories / Specialist Projects & Histology; H.O. Krueger, EAG Laboratories / Aquatic, Plant and Insect Toxicology; J.W. Green, DuPont / Data Science and Informatics

The plant interest group of SETAC has a committee working on the topic of recovery and this presentation concerns statistical issues arising from this work. Traditionally, evaluating the risk of chemicals to plant species involves assessing both lethal and nonlethal effects, but little or no consideration is given to whether the effects at the population or community level are transient or persistent. Considering the ability of plants to recover after the exposure to a chemical is important when evaluating effects on populations conducting a risk assessment. For example, a young plant in a vegetative vigor study may show leaf damage one day after exposure, but after two weeks of growth that damage may no longer be apparent as old damaged leaves have senesced and only new unaffected leaves are visible. While it is relatively easy to design studies to assess recovery of vegetative growth in terrestrial plants, this may not be indicative of recovery of the ability of a population of plants to sustain itself. In algae or lemmna studies, an aliquot of the population could be collected on a weekly basis and the ability of the algal community to recover would be assessed. In terrestrial plants it is often necessary to construct a number of mono-culture experiments and the results compared to the control.
complicated by seasonality, changes in nutrients, reclamation, competition, and other factors. Terminology and methodology need to be standardized if the concept of recovery is incorporated into evaluations of chemicals. For some plant types and properties, recovery is contingent upon the timing and duration of exposure and extent of injury. In such cases, the definition of recovery must specify timing and duration of exposure in the operational definition. Examples of recovery in laboratory studies for algae, Lemma, and Myriophyllum studies will be presented as well as results from mesocosm studies. Statistical procedures and experimental designs will be presented for these examples and interpretation of results will be discussed.

402 Aquatic primary producers and plant protection products: endpoints and level of protection achieved in the first tier of the risk assessment scheme S. Duquesnes, UBA, Federal Environment agency; L. Hömmemann, S. Matezki, M. Solé, K. Swarowsky, German Environment Agency UBA; J. Wogram, German Environment Agency UBA / Department IV plant protection products

In the risk assessment for aquatic primary producers exposed to plant protection products (PPP), the endpoint (EP) corresponding to 50% inhibition of growth (EC50) is used in the first tier. The EC50 can be expressed as inhibition of the average specific growth rate (ErC50) or as reduction in biomass, calculated from yield (EyC50) or as the integral under the growth curve (EbC50). The lowest available EP among ErC50, EbC50 or EyC50 used to be selected to derive safe concentrations of pesticides in surface water bodies. It is now recommended [1] to use ErC50 since it is a more robust endpoint. However, it is not yet clear if the protection level achieved is sufficient. This work shows that this new approach (i.e. selecting ErC50) shifts the thus of conservatism of a factor of 6.9 and 3.5 for algae and Lemma sp, respectively. It also shows that the level of protection achieved for primary producers becomes insufficient in 59% of the cases, since the Tier 3 Regulatory Acceptable Concentrations (RACs) from micro-/mesocosm studies (considered as surrogate reference Tier) are lower than the Tier 1 RACs from standard toxicity tests. The results demonstrate that the intended level of protection is currently reached in only 41% of the cases versus 69% of the cases previously. In addition, this work explores which combination of EC1o, (EC10, EC50 etc.) and assessment factor would ensure an adequate level of protection. Recommendations are provided for an optimization of the risk assessment.

403 Poster spotlight: WE152, WE153, WE154

Environmental monitoring of contaminants using terrestrial ecological biomonitor

404 Persistent Organic Pollutants in Germany: Results from the 2015/2016 moss and Odor sampling A. Dreyer, Eurofins GfA GmbH / Air Monitoring; S. Nickel, University of Vechta / 2; J. Koschorreck, Umweltbundesamt; W. Schröder, University of Vechta / 2

This presentation aims at reporting on the determination of selected POPs in eight moss samples from Germany within the framework of the 2015 European moss survey and comparing the respective results with such derived for leaves and needles collected for the German Environment Specimen Bank (ESB). The moss samples were collected in areas located in the vicinity of tree sampling sites from the ESB in fall 2016. Deciduous tree leaves and coniferous shoots were sampled in 2015 or 2016 according to the ESB protocols. Overall, 17 polychlorinated dibenzodioxins and -furans (PCDD/F), 18 polychlorinated biphenyls (PCB), 16 polycyclic aromatic hydrocarbons (PAH), 17 perfluoralkyl substances (PFAS), 3 isomers of hexabromocyclododecane (HBCD), 7 polybrominated diphenyl ethers (PBDE), 24 polybrominated diphenyl ethers (PBDE), and 21 alternative halogenated flame retardants (HFR) such as Dechlorane Plus were determined. Except for PCBs and PFASs, POPs of all substance groups could be quantified, although to different extents. Concentrations of individual PAHs and HFRs were in the same order of magnitude as those observed in coniferous shoots or deciduous tree leaves from nearby located areas. Highest levels of PCDD/F, dL-PCBs, HBCD and PAH in moss were observed at sites close to the Belauer See (Northern Germany, agricultural land-use) and the Harz National Park. Concentrations of PBDEs were highest at the two sampling sites in Saarland (conurbation) and at the Harz site. Concentrations for Dechlorane Plus were highest at the Harz site followed by sites located at Sölping (forestry) and Scheegeom (agriculture) and were lowest at the site in the Halle-Leipzig conurbation. Thus, surrounding land-use does not seem to be the (only) driving force determining the POPs burden in moss samples. PBDE moss concentrations observed in this study were similar to those observed at background sites in Spain and lower than those of background/remote sites in Norway. Concentrations of Dechlorane Plus were more than a factor of 100 higher than moss concentrations reported for Svalbard (Arctic Norway).}

Mapping percentile statistics of element concentrations in moss collected from 1990 to 2015 in forests throughout Germany W. Schröder, S. Nickel, University of Vechta / 2

Monitoring and mapping of atmospheric deposition can be achieved by use of chemical transport models, technical samppling devices and bioaccumulators such as moss. Within the European moss survey programme, since 1990 every five years moss samples from 7800 sites in 35 countries have been collected. The data, and literature derived chemical determination of heavy metals (since 1990), nitrogen (since 2005), and persistent organic pollutants (since 2010) in moss specimens, quality control and statistical evaluation was conducted according an harmonized methodology [1]. Mapping the percentile statistics of heavy metals and nitrogen concentration in moss sampled in forests across Germany is in the focus of this paper. Thereby, element- and survey-specific as well as heavy metals and surveys integrating statistical evaluations and GIS-mapping were performed. Cr, Hg, Sb and Zn show, contrary to Fe and Pb, no constant decrease of element concentrations, but an intermediate increase between 2000 and 2005, which did not continue until 2015. Al, As, Cd, Cu and V stagnated between 2000 and 2005. Hg from 2005 to 2015. Therefore, Cr, Sb and Zn will be focused in this paper together with Cd, Hg, Pb and N which are of priority according to the Convention on Long-range Transboundary Air Pollution. Survey-specific statistical analyses corroborate that the spatial patterns of element concentrations in moss are changing across time.


406 Heavy metal and nutrient concentrations in different age classes of holm oak leaves and pine needles - a reference for biomonitoring and geochemistry J. Franzaring, A. Fangmeier, University of Hohenheim / Institute of Landscape and Plant Ecology; L. Paoli, University of Siena / Dept. of Life Sciences; S. Ancora, University of Siena / Physical sciences, Earth and environment; S. Schlosser, University of Hohenheim / Core Facility Hohenheim; E. Nemanj, University of Siena / Dept. of Environmental, Earth and Physical Sciences

Passive biomonitoring is being used for many years to assess changes in the state of the environment. Existing programs make use of international, national, regional and local monitoring networks addressing the effects of the widespread deposition of air pollutants and eutrophying compounds and the accumulation of these in e.g. forest and agricultural ecosystems and their effect on the health of organisms. Recent studies showed the importance of proper food chain. However, plants differ in the uptake of essential and non-essential inorganic elements and organic pollutants and the accumulation varies over time and space mainly due to the different geochemistry and climates in different regions. Here we report on the results of a biomonitoring study using holm oak (Quercus ilex L.) and two pine species (Pinus nigra J. F. Arnold and Pinus pinaster Ait.), i.e. evergreen deciduous and coniferous respectively, to monitor heavy metal and nutrient concentrations in both leaves and needles were sampled at different locations in the Mount Amiata and Colline Metallifere region in spring 2017 and the samples consisted of three age classes. While half of the analyses were performed on unwashed samples, the other half rested on samples that were thoroughly rinsed with deionized water prior to the analyses. ICP-MS, ICP-OES and CNS elemental analyses were applied for the determination of Cu, Si, Al, Ba, Cr, Ni, V, Fe, Hg, K, Mg, As, Pb, Cd, Zn, Sn and S. Apart from the significant differences in element concentrations and patterns between the tested species, regions and sites, differences between the washed and unwashed samples was less pronounced indicating that deposition of dust does not play a great role in the area. On the other hand, our preliminary analyses showed significant differences in element concentrations between different age groups which relate to the availability, translocation, accumulation or growth limitation of plant essential and non-essential elements. Multi-element analyses and nutrient ratios can serve to differentiate between the characteristic geochemical and species-specific patterns and the positive derivation from these patterns points to the exceedance of element levels due to pollution and eutrophication. Biomonitoring in post-mining areas serves to identify pollution hotspots and can be used as a key component in controlling the success of land reclamation for agriculture and recommendations.

407 Examining historical trends in diet and contaminant exposure in bats using butyrate deposits from Jamaica L. Gallant, University of Ottawa / Department of Biology; C. Grooms, Queens University; L.E. Kimpe, University of Ottawa / Department of Biology; J.P. Smol, Queens University / Biology; W. Bogdanowicz, Museum and Institute of Zoology;
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Systemic ecotoxicology: application of OMICS data across multiple level of biological organization in research and risk assessment (I)

410 Transcriptomic responses of the endangered freshwater mussel Margaritifera margaritifera to trace metal contamination

A. Di Caprio, F. Pierson, University of Bordeaux / UMR EPOC CNRS 5805; J. Thébault, Université de Bretagne / LEMAR UMR 6539 CNRS/UO/IRD/Ifermer; C. Klop, INRA Institut National de la Recherche Agronomique / Plate-forme bio-informatique Genotoul, Mathématiques et Informatique Appliquées de Toulouse; J. Belloc, Université de Bretagne / LEMAR UMR 6539 CNRS/UO/IRD/Ifermer; P. Gonzalez, University of Bordeaux / UMR EPOC CNRS 5805; M. Baudracco, Université de Bordeaux / UMR EPOC CNRS 5805

The freshwater pearl mussel Margaritifera margaritifera is one of the most threatened freshwater bivalves worldwide. In this study, we aimed (i) to study the processes by which water quality might affect freshwater mussels in situ and (ii) to provide insights into the ecotoxicological significance of water pollution to natural populations in order to provide necessary information to enhance conservation strategies. Margaritifera specimens were sampled in two close sites located upstream or downstream from an illegal dumping site. The renal transcriptome of these animals was assembled and gene transcription determined by RNA-sequencing. Correlations between transcription levels of each single transcript and the bioaccumulation of trace metals, age (estimated by sclerochrography) and condition index were determined in order to identify genes likely to respond to a specific factor. Amongst the studied metals, Cr, Zn, Cd and Ni were the main factors correlated with transcription levels, with effects on translation, apoptosis, immune response, response to stimulus and transport pathways. However, the main factor explaining changes in gene transcription appeared to be the age of individuals with a negative correlation with the transcription of retrotransposons-related genes. To investigate this effect, mussels were classified into 3 age classes. In young, middle-aged and old animals, transcription levels were mainly explained by Cu, Zn and age, respectively. This suggests differences in the molecular responses of this species to metals during its lifetime that must be better assessed in future ecotoxicology studies.

411 L-C-HRMS based-metabolomics to highlight bionutriotrophic products and effects of diclofenac in Mytilus galloprovincialis

F. Courant, Université de Montpellier - UMR 5569 Hydrosciences / UMR Hydrosciences; B. Bonnefille, H. Fenet, Université de Montpellier / UMR Hydrosciences; E. Gomez, Université de Montpellier, Hydrosciences Montpellier UMR 5569 / UMR Hydrosciences

Diclofenac (DCF) has become a major contaminant of interest as shown by its inclusion in the EU Water Framework Directive (2015/405/EC). However, relatively little is known regarding its bionutriotropic effects and interactions in the Mediterranean mussel. Environmental metabolomics affords several advantages to study both topics. The metabolome refers essentially to i) the "endometabolome", consisting of endogenous metabolites, and to ii) the "xenometabolome", in reference to xenobiotics and their bionutriotropic products [1]. Metabolomics profiles acquired through mass spectrometric techniques may reveal the exposure by direct detection of xenobiotics and their metabolites (xenometabolite investigation) and effects by the detection of endogenous metabolites which concentrations may differ from physiological levels following the exposure. To investigate this effect, mussel specimens were classified into 3 age classes. In young, middle-aged and old animals, transcription levels were mainly explained by Cu, Zn and age, respectively. This suggests differences in the molecular responses of this species to metals during its lifetime that must be better assessed in future ecotoxicology studies.
4'-hydroxy-diclofenac, and 10 were phase II metabolites such as amino acids conjugates. Five were reported for the first time in an aquatic organism. Regarding the effects, two main metabolic pathways were found to be impacted by diclofenac exposure. The tyrosine metabolism was mostly down-modulated and the tryptophan metabolism was mostly up-modulated. To our knowledge, such DCF effects on muskels have never been described despite being of concern for these organisms. Aromatase and serotonin are involved in osmoregulation, and in gene regulation in mollusks [2-4]. Our results highlighted potential impairment of muskul osmoregulation and reproduction following a DCF exposure in agreement with recent publications that have shown reproductive disturbance following DCF exposure in other aquatic organisms such as xenopus [5] or fish [6]. 1 Holmes et al., Anal. Chem. 79, 2629 (2007) 2 Wang & Croll, Aquaculture 256, 423 (2006) 3 Fong et al., Exp. Zool. 267, 475 (1993) 4 Fong et al., J. Exp. Zool. 266, 79 (1993) 5 Efosa et al., Chemosphere 173, 69 (2017) 6 Gröner et al., Chemosphere 166, 473 (2017)

412 Metabolomics used to link affected molecular pathways with behaviour outcomes after a single dose of pesticide exposure in the polar bear
P. Leonards, VU University, Institute for Environmental Studies / Department of Environment and Health; H. Vibe, I. Lee, S. Buratovic, P. Eriksson, Uppsala University
World-wide, serious concern has arisen about the increased incidence of learning and developmental disorders in children. From a scientific point of view, there is no doubt that exposure to neurotoxic chemicals during early brain development can adversely affect learning and development. Various recent epidemiological studies have indicated that exposure to low doses of environmental biologically active contaminants (e.g. pesticides) during human development can have deleterious effects on cognitive development in childhood. The European commission-funded project DENAMIC 0Developmental Neurotoxicity Assessment of Mixtures in Child Translational Research in Human and Animal Tissue0 investigates neurotoxic effects (e.g. learning and developmental disorders) of low-concentration mixtures of pesticides and a number of common environmental pollutants in children. We focus on (sub)clinical effects on learning (cognitive skills) and developmental disorders in children (e.g. ADHD, autism spectrum disorders and anxiety disorders). The aims are to develop better and sophisticated tools, procedures and testing methods to screen compounds for (developmental) neurotoxicity, and to improve our understanding of chemical exposures and the observed effects (www.denamic-project.eu). As part of the project, a new alternative assessment strategy based on a combination of in vitro, in vivo assays, omics, and human exposure assessment is underway in order to prioritize compounds, and to further investigate the pathways and mechanisms involved in disorders and diseases. The final aim of DENAMIC is to reduce effects of environmental contamination on learning and developmental disorders in children. In the current study metabolic pathway analysis was used to improve our understanding of the underlying molecular mechanisms of observed effects on behaviour and cognitive function after various pesticide and other contaminant exposures in mice. 413 Relationships Between Persistent Pollutant and Metabolomics Profiles in Tissues of Polar Bears From Hudson Bay, Canada
A.D. Morris, Environment and Climate Change Canada / Ecotoxicology and Wildlife Health; R.J. Letcher, Environment and Climate Change Canada / Ecotoxicology and Wildlife Health Division; M. Dyck, Government Of Nunavut / Nunavut Department of the Environment; B. Chandramouli, J. Cosgrove, SGS AXYS
Metabolomics profiles are comprised of targeted endogenous metabolites (< 1 kDa) such as amino acids (AAs), fatty acids (FAs), and membrane lipids such as phosphatidylcholines (PCs) to identify how changes in the metabolism relates to extrinsic factors, including e.g. exposure to persistent organic pollutants (POPs) and metals. Polar bears (Ursus maritimus) from Hudson Bay (Canada) are differentially exposed to complex mixtures of POPs and metals including total mercury (THg = inorganic + methyl-mercury), and legacy and new POPs. In the present study, quantifiable profiles of 295 organic POPs and THg in fat and liver (representing a shift to mechanistic testing methods to screen compounds for neurotoxicity). The pooled library was used to find mechanisms of herbicide toxicity in Chlamydomonas reinhardtii to three herbicides that target photosynthesis: atrazine and diuron which target photosystem II, and which disrupts the transition of electrons from photosystem I to photosystem II, and which produces the active oxygen species. The pooled library was exposed the EC20 concentration of each herbicide for 3–4 days, at which time samples from the library were taken for DNA sequencing to find mutants that have been identified as being differently enriched or impacted between the subpopulations. Greater ARA in SHB bears may be related to differences in chronic exposure to POPs such as the hepatotoxic PFAs, as ARA is part of the inflammatory response in liver. SDMA and most legacy organochlorine compounds were greater in the WHB bears, which may be indicative of differences in renal function. Consistency of relationships between metabolites, POPs, biomarkers from laboratory studies suggests linkages between POP concentrations and differences in the hepatic metabolome of SHB and WHB polar bears.

414 Integrative Omics linkage to reproduction effects of a fungicide in the soil invertebrate Folsomia candida
T.F. Simoes, S.C. Novaes, Politecnico Institute of Leiria / MARE IPLeiria; T. Natal da Luz, University of Coimbra / Department of Life Sciences, University of Coimbra; J. Sousa, University of Coimbra / Department of Life Sciences; B. Devreese, Ghent University / Laboratory for Protein Biochemistry and Biomolecular Engineering; T. de Boer, Vrije Universiteit; D. Roelofs, Vrije Universiteit / Department of Ecological Science; N. van Straalen, Association of Renewable Environmental Scientists ARES / Department of Ecological Sciences; M. Lemos, Instituto Politécnico de Leiria / MARE IPLeiria
Due to high complexity of ecosystems, environmental risk assessment can be a challenging task and there is the need to develop and validate innovative and reliable tools and integrated approaches for fast detection of changes in population and community structures that can be applied by regulatory agencies. Folsomia candida is among the most sensitive invertebrates of its taxon, and has been selected as a genomic model organism for soil toxicology studies on non-target soil arthropods. This work aimed to determine the toxicity mechanisms of a widely applied fungicide formulation (Bravo500®), with active compound chlorothalonil (CHT), in F. candida, by linking effects at different levels of biological organization like reproduction, gene expression and protein levels, following a time series exposure. Therefore, Folsomia candida was exposed to the estimated EC50 of the formulation (plus control) and sampled at consecutive time points (2, 4, 7, and 10 days). Four replicates per treatment and time point were used (32 in total). CHT exerts its toxic effects primarily through binding to thiol-rich molecules (ex. glutathione), exhibiting often a multi-site activity and the results with the formulation were very indicative of these mechanism of toxic activity. Also in this study, results point for similar effects of the detected formulation with theechaotic mechanism of action (involving in detoxification and excretion (also involving glutathione), normal cellular respiration and protein metabolism, leading to impairment in development and reproduction. The datasets presented highly significant positive correlations between the gene expression levels at a certain time point and the correspondent protein products from the consecutive time-point, thus highlighting the importance of considering a time serie analysis when identifying potential endpoints. The exposure to POPs such as the hepatotoxic PFASs, as ARA is part of the inflammatory response in liver. SDMA and most legacy organochlorine compounds were greater in the WHB bears, which may be indicative of differences in renal function. Consistency of relationships between metabolites, POPs, biomarkers from laboratory studies suggests linkages between POP concentrations and differences in the hepatic metabolome of SHB and WHB polar bears.
stress and lipid metabolism. Finally, there were also several genes that were among the enriched separately for diuron and atrazine, which points to possible different secondary modes of action for both herbicides. While we are currently still analyzing the obtained profiles and individual genes, our results demonstrate that functional genomics is a useful method for discovery of molecular mechanisms of chemical toxicity.

**Harmful effects of plastic litter and mitigation strategies in the Mediterranean Sea**

416 **Harmful effects of plastic litter on Mediterranean Biodiversity: what and what’s new?**
M. Fossi, M. Baini, C. Panti, University of Siena / Department of Physical Sciences, Earth and Environment

Concern about the occurrence, quantity and effects of marine litter in the world’s ocean and seas has grown rapidly in recent years, attracting interest from a wide range of stakeholders, environmental NGOs, the scientific community, the media and the general public. Mediterranean Sea, which is a crucial biodiversity hotspot and a critically polluted area, has been also described as one of the areas most affected by marine litter, including microplastics, in the world. Recent studies in the different regions of the basin suggested that some areas are affected by important concentration of microplastics and plastic additives, representing a potential risk for endangered species (baleen whales, sea turtles, filter feeders sharks) and for the all Mediterranean biodiversity. To cover the current knowledge gaps on this issue a harmonised methodological approach for the assessment of the marine debris impact on Mediterranean biodiversity is needed. The quantification of marine debris/microplastics in the marine environment can depend on several environmental factors and change according to multiple oceanographic features, and therefore, cannot reflect the potential impact on organisms and ecosystems. The information obtained by biondicator species could better integrate the spatial and temporal presence of marine litter/microplastics in the marine environment. In addition, the use of biondicators can allow to measure not only the occurrence of marine litter in the species and its environment but also the threat posed to organisms by the evaluation of contaminants accumulation and any physiological/biological effect. A new integrated monitoring tool that would provide the information necessary to design and implement future mitigation actions in the Mediterranean basin is proposed. Applying ecological and biological criteria to the most threatened species obtained by statistical analysis, biondicator species for different habitats and monitoring scale were selected. To assess the harm by marine debris ingestion a threefold approach, simultaneously measuring the presence and effects (accumulation of plastic associated contaminants and biomarker responses), can provide the harm and the sub-lethal effects to organisms related marine litter ingestion. The gaps pointed out by this research and the biondicators species selected could represent a step forward for the risk assessment and the implementation of future mitigation measure for the Mediterranean area, habitat and species affected by marine litter ingestion.

417 **Impact of marine litter in the Mediterranean Sea: monitoring and specific reduction measures within MSFD**
F. Giliani, IFREMER

Periodic assessments of the state of the marine environment, monitoring and the formulation of environmental targets are perceived as part of the continuous management process within the MSFD. Of the 11 descriptors listed in Annex I of the MSFD for determining DES, Descriptor 10 has been defined as “Properties and quantities of marine litter do not cause harm to the coastal and marine environment”. In 2016, the Revised Commission Decision identified four indicators for Descriptor 10, of which two are focusing on harm considering (i) The amount of litter and micro-litter ingested by marine animals is at a level that does not adversely affect the health of the species concerned (indicator 10DC3), and (ii) The number of individuals of each species which are adversely affected due to litter, such as by entanglement, other types of injury or mortality, or health effects (indicator 10DC4). For these two indicators, Member States shall establish that list of species to be assessed and thresholds values for these levels through regional or sub regional cooperation. In the context of the Mediterranean Sea, we discuss the ongoing work that is focusing on the implementation of monitoring and reduction measures, defining constraints, protocols, better defining harm and research needs to support monitoring efforts and reduction measures. The analysis of existing data will reveal (i) the suitability of some approaches for better monitoring the adverse effects of litter, and (ii) the potential of visual observations of the sea floor for the measurement of interactions between litter and invertebrates as an approach for evaluating entanglement. Strategies for the implementation of monitoring are discussed, as well as risk assessment and the possible associated measures within MSFD.
Ecological risks under complex, multiple-stressor threat scenarios: integrating chemical effects with environmental drivers (II)

Biodiversity patterns in the GLOBAQUA basins and their relationships with multiple stressors
N. De Castro-Catalá, Universitat de Barcelona / Department of Evolutionary Biology, Ecology and Environmental Sciences; I. Muñoz, University of Barcelona / Department of Biology, Ecology and Environmental Studies; E. Kalogjimi, I. Karouzas, A. Vourka, E. Stentí, L. Vardakas, Hellenic Centre for Marine Research, Institute of Marine Biological Resources & Inland Waters (HCMR); M. Paunovic, University of Belgrade, Institute for Biological Research Sinisa Stankovic; C. Borrego, M. Petrovic, Catalan Institute for Water Research ICRA; S. Sabater, ICRA Catalan Institute for Water Research; S. Diaz, D. Baho, University of Oslo / Department of Biosciences; J. Norberg, Stockholm Resilience Centre; L. Nizzetto, NIVA Phytoplankton are crucial for lentic ecosystems productivity and foodwebs, but facing multiple antrhopic challenges that may lead to complex alterations of their ecology and function. Climate change is expected to decrease the stability of lentic ecosystems and enhance fluctuations in environmental conditions. More frequently occurring storm events will potentially disrupt the normal stratification patterns in boreal lakes, thereby dispersing algae from the depth layers they are optimally

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Management (RPML) in December 2013, providing for a set of programmes of measures and implementation timetables to prevent and reduce the adverse effects of marine litter on the marine and coastal environment. It includes innovative and traditional measures of a policy, regulatory and technical nature, addressing different aspects of marine litter prevention and management from land and sea based sources. The Regional Plan measures impose clear obligations regarding the waste management hierarchy, closure of illegal dumping/dumpsites, shift to sustainable consumption and production patterns, removal of existing marine litter using environmental sound practices e.g. fishing for litter, clean up campaigns, port reception facilities at possibly no special fees, and monitoring, assessment and reporting on implementation of measures as well as enforcement of national legislation. Significant effort has been made on marine litter at regional and national levels, since the adoption of the RPML. The Mediterranean countries have included marine litter in their updated National Action Plans (NAPs) and the ambitious and novel Integrated Monitoring and Assessment Programme (IMAP) of the Mediterranean Sea and Coast and Related Assessment Criteria has been adopted since 2016 by the Mediterranean countries including two common and one candidate indicators on marine litter. Furthermore, with the support of the EU-funded Marine Litter MED project, UN Environment/RPML is implementing key reduction and prevention measures on marine litter in the Southern Mediterranean. At the invitation of the UN Environment/MAP, the Regional Cooperation Platform on Marine Litter in the Mediterranean was established in September 2016, consisting of more than 20 international and regional partners with a clear mandate on marine litter management. The aim of the platform is to assist to the cooperation with the RPML and the Mediterranean Sea Action Plan (Marpal), a further step has been results, towards a more effective marine litter management at regional level. One of the latest developments of the UN Environment/RPML is the 2017 Mediterranean Quality Status Report (QSR) that dedicates two chapters on marine litter related to beach, floating, and seafloor marine litter.

241 Science and awareness: a Mediterranean Connection Against Marine Litter: First Results of the Commitment Presented at UN Ocean Conference G. Zampetti, Legambiente "Science and awareness: a Mediterranean connection against marine litter" is the title of the voluntary commitment that Legambiente and the University of Siena presented at the last UN Ocean Conference in New York, in June 2017. There was a connection between scientific research and raising awareness built to tackle marine litter in the Mediterranean Sea by sharing experiences and developing a new integrated action. In 2013, Legambiente started the monitoring of floating macro litter within Goletta Verde, one of the most popular campaigns of analysis and information about sea pollution. In the last few years, there has been an increase in the marine-litter-related activities, including surveys using citizen science and awareness raising projects. Following the Scientific Environmentalism purpose Legambiente applied official methods and protocols to contribute to the estimation of the marine litter amount in seas and along the coastline, cooperating also with national research institutes, universities and other research organizations. Now, thanks to the cooperation with the University of Siena, a further step has been results, carrying out studies and research on the presence of contaminants adsorbed by floating plastics and their potential effects on biodiversity. The first results of this research will be presented in this meeting.

242 Discussion

243 Final Remarks G. Leone, UNEP/Mediterranean Action Plan

245 Changes in pCO2 alter the reproductive toxicity of common active pharmaceutical ingredients C.-M. Häst, N. Wichmann, C. Lewis, K. Smith, A. Wilson McNeal, University of Exeter / Department of Biosciences; J. Snape, AstraZeneca UK Ltd. / AstraZeneca Global Environment; T.S. Galloway, University of Exeter / Biosciences Increasing pCO2 in aquatic environments is occurring as a consequence of the release of anthropogenic carbon dioxide in to the atmosphere, which is absorbed by surface waters. Traditionally this stressor has been studied in isolation, however environmental variation such as changes in pCO2 or pH can alter the ionisation and consequently the effects of contaminant compounds. A notable group of compounds susceptible to these changes include the active pharmaceutical ingredients (APIs), which often have pH-specific biological effects and are increasingly detected in sewage effluent and receiving waters. The aim of this study is to investigate the hypothesis that changes in pCO2 alter the effects of active pharmaceutical ingredients on sperm swimming parameters and fertilisation success. The species chosen to explore these effects were the lugworm Arenicola marina and the purple sea urchin Paracentrotus lividus due to them being keystone coastal species in areas where API contamination is occurring, and them being established model species for artificial spawning in controlled laboratory conditions. We used a range of neonicotinoid pesticides and non-steroidal anti-inflammatory drugs (NSAIDs) at both environmentally relevant and mechanistic concentrations to test this relationship due to them having chemical properties identified as making them pCO2-sensitive. pCO2 conditions equating to current (8.10 ± 0.1) and future (7.75 ± 0.1) pH conditions were selected for this study. Endpoints measured included a range of sperm motility parameters, using computer-assisted sperm analysis (CASA) software and fertilisation success. Our findings indicate that pCO2 conditions may play a vital role in altering the toxicity of common chemical pollutants through changes in sperm swimming parameters and consequently fertilisation success. Our results also indicate that this combination of stressors is compound-specific between contaminants with multidirectional effects dependent on conditions of the exposure. Our findings provide novel evidence that future environmental conditions may substantially alter the role which contaminants such as APIs play in the environment. API contamination is likely to increase following anthropogenic pressures such as population growth and healthcare advances. As a result, we believe it necessary to consider future conditions such as increased pCO2 conditions when accurately assessing the environmental risks of such compounds.

246 From individual traits to ecosystem functioning: natural phytoplankton community responses under combined environmental stress and chemical pollution D. Baho, Norwegian Institute for water research; E. Leu, Akvaplan-niva AS; F. Persson, Fawag Ecology Research of Aquatic Science and Technology / Aquatic Ecology; J. Moe, Norwegian Institute for Water Research (NIVA) / Section for Catchment Processes; D.O. Hessen, University of Oslo / Department of Biosciences; J. Norberg, Stockholm Resilience Centre; L. Nizzetto, NIVA Phytoplankton are crucial for lentic ecosystems productivity and foodwebs, but facing multiple anthropic challenges that may lead to complex alterations of their ecology and function. Climate change is expected to decrease the stability of lentic ecosystems and enhance fluctuations in environmental conditions. More frequently occurring storm events will potentially disrupt the normal stratification patterns in boreal lakes, thereby dispersing algae from the depth layers they are optimally
acclimated to. Many species of algae are highly adaptive and can respond by growing rapidly after disturbance. However, the diffuse burden of chemical pollutants concomitantly present in freshwater ecosystems, can favour species of algae that are more tolerant to chemical pollution to the detriment of more adaptive ones. We carried out a field experiment, using a non-invasive mesocosm approach, to study the response of phytoplankton communities to combined physical and chemical anthropogenic stressors. We aimed at investigating if chemical pollution can have a significant effect on the algal system with specific reference to extreme events. An extreme metrological event was mimicked by sampling and mixing phytoplankton communities over the entire water depth of the lake. The chemical stressors comprised of a mixture of chemical pollutants added to the mesocosms at five increasing concentrations (typically considerably below the EC40 of individual compounds). This mixture included 12 pharmaceuticals and personal care products commonly detected in lakes and rivers in Europe. In addition, a mixture isolated from treated water effluents was used as a treatment of its own. Individual level traits (cell size, pigments), community parameters (biomass, functional diversity, species composition and photosynthetic efficiency), chemical concentrations and nutrients were routinely monitored during the 38 days experiment. Overall, the simulated chemical contamination and the environmentally relevant concentrations have a drastic persistent impact on the different levels of organisation of the phytoplankton community. At community level, contaminant decreased the photosynthetic yield. At higher concentration levels these effects persisted throughout the duration of the experiment, resulting in lower productivity and communities with contrasting. This suggests that diffuse chemical stressors can disrupt the capacity of natural communities to handle environmental changes.

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The role of multiple stressors in an Alpine river and the response of the macroinvertebrate community to chemical stressors by M. Assuncao, Institute of Aquatic and Environmental Chemistry; E. Stella, University of Trento / Department of Civil, Environmental and Mechanical Engineering; E. Capri, Università Cattolica del Sacre Cuore / Institute of Agricultural and Environmental Chemistry; I. Muñoz, University of Barcelona / Department of Evolutionary Biology, Ecology and Environmental Sciences; A. Bellino, University of Trento / Department of Civil, Environmental and Mechanical Engineering

In this study the combined effects of hydrological and chemical stressors on benthic macroinvertebrates are evaluated in order to explore the response of the biological community in relation multiple pressure factors. The Adige River, located in the Southeastern Alps, was selected as a case study because representative of a variety of ecosystems. The river basin included 12 pharmaceuticals and personal care products commonly detected in lakes and rivers in Europe. In addition, a mixture isolated from treated water effluents was used as a treatment of its own. Individual level traits (cell size, pigments), community parameters (biomass, functional diversity, species composition and photosynthetic efficiency), chemical concentrations and nutrients were routinely monitored during the 38 days experiment. Overall, the simulated chemical contamination and the environmentally relevant concentrations have a drastic persistent impact on the different levels of organisation of the phytoplankton community. At community level, contaminant decreased the photosynthetic yield. At higher concentration levels these effects persisted throughout the duration of the experiment, resulting in lower productivity and communities with contrasting. This suggests that diffuse chemical stressors can disrupt the capacity of natural communities to handle environmental changes.

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Coping with antidepressants in a changing ocean: tissue bioaccumulation and behavioural implications in juvenile Argyrosomus regius exposed to venlafaxine by A. Maulvault, Instituto Português do Mar e da Atmosfera / Division of Aquaculture and Seafood Upgrading; L.H. Gabbert, Catalan Institute for Water Research ICRA; S. Rodríguez-Mozoa, Institute for Water Research (ICRA) / Water Quality; P. Anacleto, Portuguese Institute for the Sea and Atmosphere / Division of Aquaculture and Upgrading; V.F. Barbosa, Portuguese Institute of Sea and Atmosphere IPMA / Division of Aquaculture and Seafood Upgrading; R. Alves, IPMA, I.P.; J.R. Paula, Faculdade de Ciências da Universidade de Lisboa / MARE Marine Environment Center, c. rosa, MARE Marine Environment and Technology Centre; M. Diniz, UCIBIO, REQUIMTE / UCIBIO, REQUIMTE; P. Pousão, IPMA. Portuguese Institute for the Sea and Atmosphere; A. Marques, Portuguese Institute of Sea and Atmosphere IPMA / Division of Aquaculture and Upgrading

Anthropogenic activities have contributed to great environmental challenges: remarkable chemical contamination and dramatic climate change. Both factors strongly affect marine ecosystems and are expected to worsen in the future, threatening marine species’ welfare and survival. Yet, information on how fish will cope with the presence of chemical contaminants in the future is still extremely limited. Emerging and non-regulated pharmaceutical and personal care products (PPCPs) have recently become a great environmental concern, since these compounds are often discharged into the aquatic environments, but their elimination through conventional wastewater treatments is rather limited. Within pharmaceuticals of human use, venlafaxine (VFX) is one of the most ubiquitous in the aquatic environment, often reaching higher concentrations than other well-known psycho-active drugs, such as fluoxetine (i.e. Prozac). In this context, the present study was focused on assessing the first-time effects of seawater warming and acidification on VFX bioaccumulation in fish tissues, as well as the behavioural implications resulting from the exposure to these stressors (alone or combined), using juvenile meagre (Argyrosomus regius) as model organism. Overall, data evidenced that seawater temperature and pCO2 levels can strongly affect the bioaccumulation patterns of antidepressants in marine organisms. Furthermore, the distinct behavioural patterns observed when VFX contamination, acidification and warming acted alone or in combination evidenced that multiple environmental stressors should be considered when assessing fish behaviour under a future changing ocean. The results here gathered further strengthen the need to carry out greater research efforts to understand how multiple environmental stressors interact with each other.

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A modelling approach to assess present and future land use pressures on a salmonid river: a case study in the River Tamar catchment (UK) by M. Assuncao, Celas Lowestoft Laboratory; P.E. Posen, Centre for Environment Fisheries and Aquaculture Science Cefas; M.G. Hutchins, Centre for Ecology Hydrology

A linked-model approach was applied to the River Tamar catchment (Southwest, UK) to assess current and likely future impacts of land use practices on salmonid populations; Atlantic salmon (Salmo salar) and brown trout (Salmo trutta). Land use data were incorporated into a validated water quality model (QUEST) at the sub-reach and basin scale and a baseline generated for the period of 2000 to 2012. Future scenarios of water quality were also generated based on land use practices recommended under ongoing catchment initiatives. Overall, the baseline water quality parameters found to be non-compliant with “Good Status” under the Water Framework Directive, or outside the freshwater requirements for salmonids, corresponded with reported land use pressures in the Tamar, namely, catchment-wide frequent elevated levels of inorganic phosphorus and, less frequently, suspended sediments. Tested future land use scenarios bringing improvements in inorganic phosphorus levels included upgrading technology at sewage treatment plants and the implementation of riparian buffer strips, combined with a corresponding reduction in livestock density. These improvements however, were marginal therefore the tested land use scenarios should be adjusted and/or new scenarios explored. Baseline seasonal average values for water quality parameters in different areas of the catchment explained 68% of salmon and trout fry density variation, and showed how different parameters might be affecting the density of these two species. Our results suggest that catchment pressures are contributing to the regulation of salmonid fry densities in some tributaries and upper catchment reaches. Moreover, they can be used to inform local and seasonal targeted measures, aimed at improving those water parameters most influential on fry densities. These types of measures are likely to bring the highest benefits to salmonid productivity in the catchment.

PBt/PvB & PMT/PvM substances and Non-extractable residues (NER): Scientific strategies, Analytical challenges and Regulatory Issues (II)

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Evaluation of PBt and PvB substances based on exposure dynamics, use-specific impacts and costs for emission reduction or abatement in the context of REACH by S. Sjöström, Wageningen University / Social Sciences; F. Oosterhuis, Vrije Universiteit Amsterdam / Institute for Environmental Studies; S. Hahn, Fraunhofer ITEM / Chemical Risk Assessment; M. Klein, Fraunhofer Institute for Molecular Biology and Applied Ecology IME; M. Nendza, Analytisches Laboratorium

A key of the European chemicals legislation REACH is to ensure that the risks caused by substances of very high concern (SVHC) are adequately controlled. The two regulatory procedures adopted in REACH to control the risks arising from SVHCs are restriction and authorisation. Both regulatory instruments make use of socio-economic analysis (SEA), which is generally defined to be a tool for assessing all relevant positive and negative impacts from substances’ use or non-use, and for comparing these impacts across different scenarios. Impacts arising from chemicals’ use, including PBt/PvB substances, are use-specific. Furthermore, due to stock pollution properties of PBt/PvB substances, impacts may last for long periods and even long after emissions have ceased. In addition, information about (long-term) impacts needs to be balanced with costs of emission reduction and abatement. Acknowledging that monetary valuation of impacts using stated or revealed preference methods is not possible for a broader set of PBt/PvB substances, the evaluation of PBt/PvB substances in a SEA has to rely on cost-effectiveness analysis (CEA). This requires specifying benchmark values, i.e.
target values (standards) by means of which it can be determined whether or not the costs of a control measure are excessive. This paper suggests an approach for the evaluation of PBT/vPvB substances by means of CEA that accounts for the complex environmental distribution patterns, and that allows balancing (long-term) impacts from PBT/vPvB use against costs for emission reduction and abatement. The approach proceeds along a sequence of steps and uses different analytic tools and data. Starting with a grouping and ranking of PBT/vPvB substances, exposure dynamics are analyzed with a multimedia fate and transport approach. Based on the assessment of exposure dynamics in different compartments, impacts arising from the stock can be evaluated via different routes. To assess the cost-effectiveness and proportionality of possible (policy-)measures for PBT/vPvB control, the routes to impact evaluation are linked to assessments of costs for restricting or stopping a specific use or multiple uses of a PBT/vPvB substance, and to benchmarks being standard values of a specific parameter to which the actual/estimated value of that parameter can be compared. As illustrative case study, the approach is applied to perfluorocarbox sulfonate (PFOS).

431 Grouping and relative ranking of the impact potential of PBT/vPvB substances for comparative assessments in the context of socio-economic analysis under REACH

M. Nedzka, Analytisches Laboratorium; S. Hahn, Fraunhofer IPEM / Chemical Risk Assessment; M. Klein, Fraunhofer Institute for Molecular Biology and Applied Ecology IME; F. Oosterhuis, Vrije Universiteit Amsterdam / Institute for Environmental Studies; S. Trapp, Technical University of Denmark / DTU Risø National Laboratory. The assessment of PBT/vPvB substances under REACH may evaluate long-term exposure dynamics and impact potential. Grouping and relative ranking of PBT/vPvB substances can support comparative assessments of either several substances with the same use, or for a particular substance with different uses (emission patterns) in the context of socio-economic analysis (SEA). The aim of the grouping and ranking is to establish a multimedia chemical with respect to its similarity of properties/behaviour with chemicals with known impacts, serving as points of reference for the impact evaluation. The relative ranking of PBT/vPvB substances is based on an impact score, which captures diverse properties and effects of target substances. The impact score consists of individual scores assigned to expected environmental stocks, possible effects of PBT/vPvB on the environment and on human health via the environment (toxicological benchmarks), and to other specific concerns. The resulting fingerprints of concern (pattern of the individual scores) and the overall scores for impact potential can be used for comparative assessments. For example, if similar fingerprints are observed for two substances with a similar use, the total score gives an indication which substance might be less (or more) critical. For 17 case study chemicals, data on partitioning properties (log Kow, log Koa), stocks in water/sediment and soil, long range transport potential (LRTP), overall persistence (Pov), toxicological benchmark values for water and soil, CMR properties (H-phrases), endocrine disruption (ED) potential, production/emission volume, and use pattern have been collected from REACH dossiers or estimated with suitable tools. The examples illustrate the possibility of multiple uses of a PBT/vPvB substance, and to benchmarks being standard values of a specific parameter to which the actual/estimated value of that parameter can be compared. As illustrative case study, the approach is applied to perfluorocarbox sulfonate (PFOS).

432 Interpretation of non-extractable residues (NERs) in the persistence assessment

U. Jöhncke, Federal Environment Agency (UBA) / IV 2.3 Chemicals; V. Bonnomet, European Chemicals Agency / ECHA; I. Doyle, Environment Agency / Evidence Directorate; R. Hornek-Gautscher, Environment Agency Austria; A. Kapanen, European Chemicals Agency - ECHA; M. Kästner, Helmholtz centre for environmental research - UFZ; K. Rege et al., Environment Agency Austria; J.R. Peltola-Thijs, ECHA-European Chemicals Agency; L. Ribeiro; A. Schäffer, Institute for Environmental Research RWTH Aachen University; M. Telscher, Bayer AG Division CropScience/Environmental Fate / Development Environmental Safety; F. Schmidt, C. Leake, Bayer AG Non extractable residues (NER) so called “bound residues” of plant protection products are formed in soil as a result of degradation processes. Due to their inherent nature, analysis and further assessments of bound residues are challenging. In a recent publication (Possberg et al. 2016) a distinct analysis of NER has been comprehensively discussed. Furthermore, different soil matrix destabilising and destroying procedures are evaluated in order to characterise the four NER types proposed by Eschenbach et al. [2]. Finally, a refined extraction scheme shall be proposed with respect to the general applicability for an adequate risk assessment of NER. [1] Kästner et al. 2014. Classification and Modelling of Nonextractable Residue (NER) Formation of Plant Protection Products. In: Soil - A Synthesis. CFeII:1; 512 pages. The method relies on the determination of natural amino acids as the main part of biogenic residues in soil. The amino acids were liberated via a digestion of the soil with 6 N HCl at 110°C. Within this presentation we focus on the utility and validity of this analytical method using 14C bromoxynil and an agricultural soil from Germany. As a main result about 55% of the labelled residues could not be liberated and remained bound to the soil even after such a harsh digestion step. During further clean-up of amino acids further losses of radioactivity of approximately 40% of those liberated bound residues has been observed. Further analyses elucidated up to 50% of those unidentified losses, however, in total approximately 75% of bound residues stuck to the soil and therefore could not be identified or unambiguously assigned. However, 16% of the generated NER was extractable and could be assigned to amino acids. Both results indicate that bound residues will not contribute to future risks or hazards because they are on one hand not bioavailable or on the other hand they have been converted to natural biogenic residues like natural amino acids. 435 A tool to establish the role of Non-Extractable Residue (NER) in soil on toxicity J. Harmsen, Wageningen University / CALM; D. Hennenke,
C. Chau, University College London / Department of Biochemical Engineering; N. Titchener-Hooker, University College London / Department of Biochemical Engineering; P. Lettieri, University College London / Chemical Engineering

6-Aminopenicillanic acid (6-APA) is the beta-lactam nuclei of penicillin and is the intermediate to most semisynthetic antibiotics. Manufacturing of the nuclei represents one of the largest production scale processes within the biopharmaceutical industry. The energy mix associated with the industry are poorly understood, due to limited life cycle assessment (LCA) studies in the literature, the paper presents a LCA of 6-APA production to illustrate the burdens manufacture places on the environment as a function of manufacturing location. We make recommendations for future development of large scale biopharmaceutical manufacture by drawing on our 6-APA analysis where necessary. With the assumed energy mix to a Brazil mix (constituting a higher proportion of renewable resources), the contribution to climate change was reduced by 15%. Manufacture in China and India where coal combustion is the main source for electricity; the emissions were significantly higher (20%). Other location dependent variables were inputted into the model in combination with the switch of energy mix. Depending on the location’s water scarcity, the burden of 6-APA on this element varies greatly, a case in point is the industry that occurred with Carbendazim. This gave a very large uncertainty about NER using non-labelled compounds. For these compounds it is not possible to draw conclusions from only a non-labelled experiments.

LCA and beyond - integrating sustainability and/or other dimensions to improve decision support (I)

436 How to make LCA fit for purpose as decision making tool
E. Mieras, PRe Sustainability; A. Gaasbeek, PRe Consultants / Consultancy; J. Courtois, PRe Consultants

To understand how Life Cycle Thinking (LCT) and Life Cycle Assessment (LCA) can be improved to support decision making we first have to look at how decisions are made. One of the first distinctions that are made when it comes to decision making is between normative and descriptive decision making. The latter describes how people actually make decisions and that can be quite irrational, as also Daniel Kahnemann, the Nobel Prize Winner, argues in his book “Thinking Fast and Slow”. The first tries to define how to come to the best option, assuming that decision makers are fully rational and that the world can be modeled as a known set of parameters. The approach of Ortega-Calvo et al. (2015) has been followed, because this approach defines clear and measurable fractions. The only not measurable fraction is NER, but can be considered as a residual fraction if all others are measured. Considered are: Chemical present in the water phase, actual available, (Passive sampling or CaCl₂-extraction) A potentially available fraction in equilibrium with the water phase (TEN Box TS -16751); The total extractable amount, measured with (a standard) method NER is considered, but mentioned as non-measurable and also non-bioavailable. We studied three NER-forming chemicals and followed their fate in a period of 6 months after addition. An important part of the study were experiments using ¹³C chemicals. In first experiments formation of Non-Extractable ¹³C was observed for all chemicals. For the chemical Tri-NitroToluene (TNT), NER-formation was reproducible and NER formation during aging removed toxicity. By removing the bioavailable fractions directly after spiking and after aging it was also possible to remove toxicity. The experiments with and also without labelled TNT clearly showed that toxicity was caused by the bioavailable chemical and not by NER. The tool developed can be used if the fate of the chemical including NER formation is obvious Wih the other selected chemicals Cypermethrin and Carbendazim, results were less clear, because there was a large uncertainty in NERformation. The degree of biodegradation was not reproducible for Cypermethrin and unexpected losses occurred with Carbendazim. This gave a very large uncertainty about NER using non-labelled compounds. For these compounds it is not possible to draw conclusions from only a non-labelled experiments.
to be used, including LCA, but not necessarily limited to it. Moreover, in every LCA, it is in a first step important to specify goal and scope for the further analysis, and it is worthwhile to be aware of aspects which have an influence on the overall environmental impacts of an investigated product. So far, goal and scope in LCA is conducted typically without a diagram or visualization of relations between different aspects to be decided about in goal and scope. We introduce influence diagrams and advanced hot spot analysis, a method to both "tailor" the approaches to be applied for dynamic LCA, the sustainability of a given situation. Both influencing variables and temporal scope of an LCA, where LCA is part of said portfolio. As an application, we develop and present a causal loop diagram for sustainability assessment of mining in general, and apply this to specific mine sites in Finland, Portugal, and South Africa, where this approach is currently applied, led by GreenDelta, in the European H2020 research project ITERAMS. In the presentation, the developed causal loop diagram and the approach for obtaining the diagram for the case will be explained, with results from the ITERAMS project. Results are quite promising and we believe that using causal loop diagrams in sustainability and life cycle assessments helps to clarify selection of the (combination of) appropriate tools for the assessment, and help to structure the goal and scope setting in LCA.

440 Life Cycle Sustainability Assessment for Improved Space Mission Design A.R. Wilson, M. Vasile, University of Strathclyde / Department of Mechanical & Aerospace Engineering; K. Baker, Glasgow Caledonian University / School of Engineering & Built Environment

The adoption of the Paris Agreement and Sustainable Development Goals in 2015 has been the driver for a more coordinated global approach towards achieving environmental sustainability. However, to be successful, this vision must run through every sector of society and the space industry is no exception. In the context of renewed global awareness on environmental sustainability, Life Cycle Assessment (LCA) is an important environmental management technique introducing a whole-systems approach within the space industry to assess the environmental impacts of products over their entire life cycle. The European Space Agency (ESA) began work on this topic in 2009, employing an internal concurrent design study called ECOSAT to consider the life cycle impact of the design, manufacturing, launch and operations of a satellite. Since then ESA has continued to develop LCA methodology for the space sector, creating the first set of LCA guidelines for space systems in 2016 and now in progress to integrate LCA into the concurrent design process. Whilst space-based LCA is still in its early stages, its further development relies on it being increasingly employed within the broader space sector to give parity across the industries. For this reason, moving towards space-based Life Cycle Sustainability Assessment (LCSA) is a logical next step which allows for the third pillars of sustainability (environment, society, economy) to be addressed within one assessment. Tailoring this integration for space systems will allow the industry to become more accountable and responsible for their operations by taking into account the full spectrum of life cycle sustainability issues associated with the operation of space systems. This paper will present the LCSA methodology used in an open-source platform under development at the University of Strathclyde, outlining the technical and economic aspects with environmental LCA to evaluate the life cycle impacts of space systems. As adverse impacts are more difficult to modify the later into the design process that they are identified, the integration of LCSA into the concurrent design process is essential for the early mitigation of sustainability issues. As such, the intention of this platform is to help decision-makers choose sustainable technologies and products at the design stage by determining those that are not only cost-efficient, eco-efficient or socially responsible, but also ones that can easily justify and evidence their sustainability.

441 How can Agent-based Modeling improve decision making in Life Cycle Assessment? A.M. Peterson, University of Bordeaux / The Life Cycle Group CyVi; P. Loubet, University of Bordeaux / ISM CyVi; F. Taillandier, University of Bordeaux / I2M GCE; G. Sonnenmann, Universität Bordeaux / ISM CyVi

Life cycle assessment (LCA) is now acknowledged as the worthiest methodology to evaluate environmental performance of whole system in a holistic way. Thus, it has been tempting to extend LCA to support both environmental management technique, including biostats, limit values of some contaminants should be fixed to consider ESR as by-products, the allowed treatments as normal industrial practice should be clearly stated. Current legislation may have a non-essential impact on the overall execution time. In this paper the legislation framework of Italy, United Kingdom and United States will be discussed. The new circular economy package of the European Commission will push all the member states to move from linear to circular economy with consequent reduction of natural resource exploitation. ESR can be one of the important sector where the challenging objectives may be reached. Now in Europe does not exist a clear understanding of the legislative measures and technical rules needed to harmonize the ESR management. Protocols are needed for their characterisations, included biostats, limit values of some contaminants should be harmonized. In this context, the Northern part of Italy authorities have allowed to use the ESR as by-products while in the area of Rome management as waste prevails. Other cases such as Crossrail (the railway tunnel crossing London), Citryingen, (Copenhagen underground) and Le Grand Paris (Paris metro) will be discussed. The new circular economy package of the European Commission will push all the member states to move from linear to circular economy with consequent reduction of natural resource exploitation. ESR can be one of the important sector where the challenging objectives may be reached. Now in Europe does not exist a clear understanding of the legislative measures and technical rules needed to harmonize the ESR management. Protocols are needed for their characterisations, included biostats, limit values of some contaminants should be fixed to consider ESR as by-products, the allowed treatments as normal industrial practice should be clearly stated. Current legislation may have a non-essential interpretation and therefore the operators are exposed to uncertainties. The general principles of fair competition inside Europe are totally disregarded.

442 Characterization and management of excavated soil and rock G. Mininni, CNRIRSA; A. Sciotti, F. Martelli, Italferr SpA

This paper describes some case studies relevant to management of excavated soil and rock (ESR) produced when tunnelling is performed using Tunnel Boring Machine with Earth Pressure Balance system (EPBs-TBM). ESR can be alternatively qualified as waste or as by-products depending on utilization conditions (in particular, certainly of further use) and on their characteristics (especially in terms of environmental and health protection requirements). Tunnels construction entails production of large quantities of ESR, up to several millions of tons. Their management as a waste certainly requires a huge amount of financial and environmental impact. A future management of ESR can be done for soil treatment to be used, including LCA, but not necessarily limited to it. Moreover, in every LCA, it is in a first step important to specify goal and scope for the further analysis, and it is worthwhile to be aware of aspects which have an influence on the overall environmental impacts of an investigated product. So far, goal and scope in LCA is conducted typically without a diagram or visualization of relations between different aspects to be decided about in goal and scope. We introduce influence diagrams and advanced hot spot analysis, a method to both "tailor" the approaches to be applied for dynamic LCA, the sustainability of a given situation. Both influencing variables and temporal scope of an LCA, where LCA is part of said portfolio. As an application, we develop and present a causal loop diagram for sustainability assessment of mining in general, and apply this to specific mine sites in Finland, Portugal, and South Africa, where this approach is currently applied, led by GreenDelta, in the European H2020 research project ITERAMS. In the presentation, the developed causal loop diagram and the approach for obtaining the diagram for the case will be explained, with results from the ITERAMS project. Results are quite promising and we believe that using causal loop diagrams in sustainability and life cycle assessments helps to clarify selection of the (combination of) appropriate tools for the assessment, and help to structure the goal and scope setting in LCA.

Environmental risk assessment and management of the material produced in tunnelling excavation

443 REALIZATION OF ROAD GALLERY: ADVANTAGES, CRITICALITY AND FUTURE PERSPECTIVES A. Selleri, Autostrade per Italia / direzione tecnica; S. Frisiani, Spea Engineering S.p.A.

Those who carry out public works at the service of the territory, which are the motorway routes, it is essential that in all phases, from approval to the realization of the work, the respect of the pre-established times in the project and the relevant regulations is guaranteed. In fact, time is a factor that directly affects the costs of execution and, above all, the costs borne by the community, which, in the event of bureaucratic red tape, cannot benefit from the availability of a more efficient, safer and less impactful service. Even though the regulatory framework tends to introduce simplifications in procedures with the latest updates, there are rare cases where there are no burdens that often negate the positive effects of investments in technologies put in place to improve construction techniques, and increase the speed of completion. To the complexity inherent in the realization of linear works, which involve the excavation of tunnels and the consequent management of the excavated soil, is added the paradox that, at equal environmental conditions, the same lands can be considered by-products or waste, even if deriving from the same pile and if produced with the same excavation system within the same work. A case that represents this situation well is the mechanized excavation technique, whose adoption often involves an excess of provisions that substantially increase the size of the characterization areas inside the building sites, and the observation times, against a null environmental advantage.

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Management of the spoil material produced by EPB-TBM: from experimental design to the excavation phase

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The use of foaming agents and additives is one of the fundamental factors allowing the correct use of the EPB-TBM (Earth Pressure Balance-Tunnel Boring Machine) for the excavation of underground works. On the other hand, their use must be carefully assessed in environmental terms, starting from the initial planning stages, in order to meet the legal requirements and, above all, avoid the subsequent use of spoil materials not pose risks for the environment and human health. During the environmental design of the project, it is therefore essential the developing of a site-specific conditioning study for the management of excavation material. In fact, on the basis of the results obtained from the conditioning tests, it is possible to hypothesize a maximum dose of the required conditioning agents, which will constitute the reference for the environmental risk assessment in the context of a sustainable management of spoil materials. Anyway, the management of spoil materials produced from excavation by EPB-TBM is extremely complex and is based on numerous design assumptions, characterized by high levels of uncertainty: from the definition of the commercial products to be used and of the most suitable soil conditioning parameters, to the execution of analysis and tests to be performed (biodegradation and/or ecotoxicological), up to the methods of interpreting the results. The uncertainties connected to the experiments carried out during the design phase can be reflected in: differences in the operational protocols to be adopted during construction, unavailability of the approved project, consequent increase in time and costs to reiterate the site-specific experiments and the failure to carry out the necessary tests. The environmental legislation on this field, clarifies only partially how to control and manage the substances contained in foaming agents and additives used for the excavation process by EPB-TBM. To overcome the design problem related to the interpretation or absence of regulatory reference limit values for the main substances contained in the additives, site-specific experiments were carried out in some projects, aimed at evaluating the eco-compatibility of the conditioned soils through biodegradation and ecotoxicological studies of the foaming agents.

445 Environmental effect of chemicals injected into the soil in mechanized tunnelling applications

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In recent years the management of the soils and rocks resulting from the mechanized tunnel excavation process, raised increasing concern in Europe. The need of a correct amount of chemicals used to treat soil, to reach a satisfactory workability of the excavated material has lead companies, governments and research groups to face economical, technical, logistical and environmental issues. The most widely used technique in mechanized tunnelling, named Earth Pressure Balance (EPB), requires the continuous injection of chemicals during the advance of the machines to enhance the excavation activity. Depending on the excavated materials features, vapours or foams can be possibly used. The environmental legislation on this field, clarifies only partially how to control and manage the substances contained in foaming agents and additives used for the excavation process by EPB-TBM. To overcome the design problem related to the interpretation or absence of regulatory reference limit values for the main substances contained in the additives, site-specific experiments were carried out in some projects, aimed at evaluating the eco-compatibility of the conditioned soils through biodegradation and ecotoxicological studies of the foaming agents.

The increasing use of Earth Pressure Balanced Shields (EPB-TBMs) in the tunnelling industry has been due to their advantages over conventional excavation methods, such as continuous operation, safer working conditions, reduced damage at surface level and higher tunnelling speed. The performance of EPB-TBMs relies on the use of appropriate soil conditioning foaming agents containing water solutions of surfactants, mainly sodium laurel ethyl sulphate (SLES) and in smaller concentrations other additives. In accordance with the Italian legislation, spoil material from excavation processes can be re-used by-products if the chemical thresholds for organic and inorganic contaminants (e.g. heavy metals, hydrocarbons C12; Italian Decree 120/2017) are not exceeded. However, there are currently neither SLES soil threshold limits in European and Italian legislation (Annex 4 of the Italian Decree 120/2017) , nor comprehensive studies on its possible ecotoxicological effects on soil and water organisms. The ecological approach here reported, consisting of site-specific studies together with ecotoxicological tests performed on the real excavated soils, aims to fill the gap between the lack of threshold limits in soil and water for these multicomponent commercial products and the environmental protection. The studies are planned following a preliminary evaluation of the foaming agent treatment ratios to be used for the specific lithological characteristics of the excavated soils. Here we report the main steps of the environmental studies useful for producing a “Protocol for the assessment of environmental compatibility of the spoil material during the tunnelling in the construction site”. The aim of the protocol is to address engineering contractors and stakeholders (e.g. Railway and Motorway operators) on how to verify the environmental compatibility of excavated soil before putting it in the destination site. It is very important to highlight that the protocol (e.g. the ecotoxicological test selection) has taken into account the site-specific characteristics and the possible environmental exposure scenarios in order to protect ecosystems and human health.

447 Mineral-based soil conditioner for EPB TBMs: An environmentally friendly alternative

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A novel product, based on a natural mineral has been developed for use as a foaming agent and soil conditioning agent with earth pressure balance (EPB) tunnel boring machines (TBMs). It is available as readily water dispersible granules or pre-prepared aqueous suspension form and can be dispersed and diluted readily in water with low shear mixing. The resulting dilute suspension can be converted into a foam using an industrial foam generator, or by other mixing methods. It can be used with existing equipment found on EPB TBMs, without the need for further modifications and investments. The use of this product to stabilise spoil material during EPB tunnelling is considered environmentally friendly because the natural mineral is widely distributed in the earth’s crust. It is virtually insoluble in water and has no known ecotoxicity. Specifically, there is an absence of toxic effects on two aquatic organisms (Danio rerio and Daphnia magna) and a demonstrated low risk to arthropods, earthworms and soil bacteria. In a recent study commissioned with an environmental consultancy, it was considered that excavated soil, treated with the product and deposited in a licenced waste facility would not pose a risk to the surrounding water environment. Compared with existing products it contains very low levels of synthetic chemicals, relying instead on the natural mineral component to stabilise foams by a completely different mechanism; one that is potentially less susceptible to the degrading influence of soils that can compete for the surfactants that are present in, and that stabilise conventional foaming tests at an independent development laboratory have demonstrated the good stability of the foams produced using the product (half-life measurements of water drainage), and confirmed that mixtures of the foam with fine and coarse grained soils have the desired consistency and cone slump behaviour for EPB tunnelling. A series of specifically developed tests reveal that the addition of foam substantially reduces adhesion of the finer grained soils to metal surfaces. In practice this will translate to a substantial reduction in the clogging potential of excavated clay in the TBM cutter head and spoil conveyors, crucial for maintaining good advance rates. The new product will be attractive to those seeking to minimise the environmental impact of tunneling projects.

Analysis and Fate of Emerging Contaminants in soils, water and plants under water scarcity (II)

448 Quantification of Carbon Nanotubes in Complex Matrices: Possibilities of Electron Microscopy

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Fascinating properties of Carbon nanotubes (CNTs) allow the development of novel materials with increased functionalities (e.g. reduced weight, increased strength). Nevertheless, CNTs do pose potential environmental and human health risks and reliable methods to quantify CNTs at low concentration in complex matrices are still lacking. A novel development laboratory based on a unique shape of the CNTs to quantify these materials in complex matrices. Multiwalled CNTs (IRMM 382) suspended in either ultrahigh quality (UHQ) water or in soil
extracts were directly centrifuged on transmission electron microscopy (TEM) grids, resulting in an even distribution of the CNTs on the grids. Samples were investigated with a scanning electron microscope (SEM, Magellan XHR 400, FEI) operated at an acceleration voltage of 20 kV in emersion mode and using a bright field transmission electron detector. A ridge detection algorithm implemented in the image processing software Fiji was used to detect and characterize individual CNTs. The bioaccumulation of CNTs was calculated based on the total length of all CNTs (provided by the ridge detection algorithm) detected on the images in combination with their thickness (20 nm), their density (1.4 g cm\(^{-3}\)) and the well-defined volume of suspension that was centrifuged on the TEM grids. CNTs were well separated on the TEM grids and an increasing number of CNTs was observed on images with increasing concentrations of CNTs in suspension. Plotting the calculated concentration of CNTs in suspension against the nominal CNT concentrations (10 \(\mu\)g L\(^{-1}\) - 100 \(\mu\)g L\(^{-1}\)) resulted in a linear relationship. The calculated nominal CNT concentrations were in good agreement at low CNT concentrations, but at high concentrations, the calculated concentrations underestimated the nominal values by a factor of ~2. Almost identical results were obtained from CNTs in UHQ water and in soil extracts (5 mg g\(^{-1}\)) indicating that the detection of the CNTs was not compromised by the presence of soil particles. Future experiments will focus on a selective removal of the soil particles by an additional treatment with diluted hydrofluoric acid. Initial experiments are promising and suggest that the detection limit of the methods can be lowered to 1 mg CNT)/kg soil), which would represent huge step forward in detecting of CNTs in complex matrices.

459 Monitoring for perfluorinated compounds, insecticides, and brominated flame retardants in the water of Daechung lake and Geum river basin

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450 Impacts of Contaminants of Emerging Concern on Terrestrial Organisms

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Reclaimed water is a historically underutilized resource. However, with increased population growth and global climate change placing increased pressure on fresh water resources, there is evolving recognition of reclaimed water as a sustainable means to meet the needs of citizens, industries, and agriculture. The use of recycled water for agriculture comes with the potential risk of environmental and food contamination by contaminants of emerging concern (CECs). These compounds pose a potential threat to the health of ecosystems because they are designed to be biologically active at low concentrations and are considered “pseudo-persistent” due to their chemical stability and low biodegradation rate. For example, pharmaceuticals and metabolites could be thousand times more concentrated in fish than in polluted living waters. By other hand, fish are known to possess a hepatic detoxification system which are likely capable of metabolizing pharmaceuticals taken up from polluted waters. Some studies proposed the analysis of bile from fish to evaluate pharmaceutical exposure including the identification of metabolites by UPLC-HRMS. In this context, we propose the evaluation of the metabolism of frequently detected fish in fish, performing a screening bile are used for the presence of stable intermediates. Fish were collected from different regions in the Mediterranean coast of Spain. Afterwards, their bile was isolated from the fish and analyzed for the detection of parent drugs and some metabolites listed in an in-house suspected list. A sampling campaign was planned to collect tuna fish, Euthynnus alletteratus, from the Mediterranean coast of Spain. Tarragona, Barcelona, and Mallorca were selected as candidate sites, every two months (March to December). In total, 17 bile samples were analyzed directly by UPLC-HRMS after a protein precipitation. The HRMS data allowed screening for suspected pharmaceuticals and their metabolites and provided plausible chemical formulae. The comparison of MS/MS spectra of the parent compounds and their metabolites allowed to propose chemical structures for possible metabolites in fish bile. With this analytical methodology some metabolites, corresponding to different reactions that includes products of hydroxylation, glucuronide conjugates were identified. The suspect analysis of bile samples allowed the detection of several pharmaceuticals. Psycho-active drugs were one of the most commonly detected drugs. Their identities were proposed by matching their accurate MS and MS/MS data against different libraries. Finally, authentic standards were employed to confirm the proposed drug identities and to determine analyze concentrations in the fish samples.

452 Accumulation and fate of 12 human drugs through the soil-root-leaf system

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CROP irrigation with reclaimed water has become an extended practice in many countries worldwide where the water scarcity and excessive exploitation of agriculture are forcing local authorities to look for alternative sources. Despite this, there are increasing levels of herbicides and contributes to nutrient recycling. Using reclaimed water for irrigation, however, represents a primary source of emerging organic contaminants resistant to wastewater treatment processes, such as pharmaceuticals and personal care products [1]. These pollutants can be retained in the soil, directly uptaken by crops or translocated from soil to plant tissues above the ground [2,3]. The present work aimed to evaluate the transfer and the bioaccumulation of organic contaminants of emerging concern (mainly pharmaceuticals) in lettuce tissues and soil. The distribution of twelve relevant wastewater-derived pollutants was evaluated in lettuce tissues (leaves and root system) and soil. This list included nine prescription drugs (dichofenac, trimethoprim, carbamazepine, oxcarbazepine, lamotrigine, cisdiltiazem, valsartan, midazolam, methadone), an illegal drug (cocaine) and two transformation products (acidozide and valsartan acid). Lettuce plants were grown in pots in a controlled environment and irrigated with artificial spiked water containing the 12 compounds during the entire growing period (60 days). Control was irrigated with tap water. Afterwards, a set of new lettuce plants were grown in the same soil pots and irrigated with rainwater or with tap water, if necessary. At the end of each growing season, leaves and root system were collected to the end of the growing season. Scanning electron microscopy. In this context, we propose the evaluation of the metabolism of frequently detected fish in fish, performing a screening bile are used for the presence of stable intermediates. Fish were collected from different regions in the Mediterranean coast of Spain. Afterwards, their bile was isolated from the fish and analyzed for the detection of parent drugs and some metabolites listed in an in-house suspected list. A sampling campaign was planned to collect tuna fish, Euthynnus alletteratus, from the Mediterranean coast of Spain. Tarragona, Barcelona, and Mallorca were selected as candidate sites, every two months (March to December). In total, 17 bile samples were analyzed directly by UPLC-HRMS after a protein precipitation. The HRMS data allowed screening for suspected pharmaceuticals and their metabolites and provided plausible chemical formulae. The comparison of MS/MS spectra of the parent compounds and their metabolites allowed to propose chemical structures for possible metabolites in fish bile. With this analytical methodology some metabolites, corresponding to different reactions that includes products of hydroxylation, glucuronide conjugates were identified. The suspect analysis of bile samples allowed the detection of several pharmaceuticals. Psycho-active drugs were one of the most commonly detected drugs. Their identities were proposed by matching their accurate MS and MS/MS data against different libraries. Finally, authentic standards were employed to confirm the proposed drug identities and to determine analyze concentrations in the fish samples.
453 Root-uptake and dissipation of atenolol, sulfamethoxazole and carbamazepine applied as a single compound solution or in mixture of all compounds in three soils and five plants

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This study was focused on a root uptake of carbamazepine, atenolol and sulfamethoxazole from 3 soils: Haplic Chernozem, Haplic Cambisol and Arenosol Epiauvtic. Five plants (radish, arugula, lettuce, spinach and green peas) planted in those soil were initially irrigated by fresh water and next with water contaminated by a single compound or their mixture. After 3 or 4 weeks, each plant was divided into separate parts: roots (including bulbs of radish), leaves, stems (green peas) and pods (green peas). Plant parts and soils were freeze-dried and dry-masses and concentrations of pharmaceuticals and their metabolites were measured. Despite that atenolol and sulfamethoxazole relatively rapidly dissipate from soils, they and metabolites of atenolol were detected in all plants. Carbamazepine is very stable in soils and fractions of its metabolites are usually low. However, very high concentrations in all plants were measured not only for carbamazepine but also for its metabolites. The degree of compounds’ transformation depended on a plant family. Considerably higher concentrations of atenolol, sulfamethoxazole and metabolites of atenolol were measured in roots in comparison to those in leaves and soils. In the case of carbamazepine, the highest concentrations were measured in leaves followed by roots and soils. Both indicate a high potential of plants to accumulate studied pharmaceuticals in their bodies and a high ability to transform studied compounds. Particularly in the case of carbamazepine, the considerably higher concentrations of metabolites were measured in leaves in comparison to concentrations in roots and very low or negligible concentrations in soils. Transformation of compounds in plant bodies is attributed to enzymes CYP450. Larger concentrations of carbamazepine metabolites were measured in leaves of lettuce, spinach and green peas than in leaves of radish and arugula (Order – Brassicales, Family – Brassicaceae). Oxcarbazepine was detected only in plants (mixture of five plants) treated as type of mixture. The lowest concentrations were measured for all tested plants. The impact of application (single compound versus compounds’ mixture) differed for different plants. Antibiotic sulfamethoxazole likely reduced dissipation of other two compounds in soils, which increased relative concentrations of compounds in plants (i.e., concentrations of compound in plant divided by compound loads in soils that is a total amount of applied solution divided by a dry mass of soil).

Prioritisation and Intelligent Testing of Pharmaceuticals in the Environment (II)

454 Inter-individual variation in the bioavailability and effects of NSAIDs in fish

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A basic tenet in the environmental risk assessment of pharmaceuticals is that pharmacological effects will occur in advance of any adverse effects, if their molecular targets are conserved in wildlife and if circulating blood plasma concentrations approach therapeutic concentrations established in humans. Fish generally display high levels of conservation of human drug targets and may be exposed to pharmaceuticals via discharges from wastewater treatment plants. The Non-Steroidal Anti-inflammatory Drugs (NSAIDs) ibuprofen and diclofenac are present in effluents, resulting in low mg/L concentrations in surface waters and fish blood plasma below or bordering on “therapeutic” concentrations. However, some studies suggest that diclofenac and ibuprofen can induce harmful effects in fish at measured environmental concentrations. Here we seek to refine the estimates of the individual variation of NSAIDs by gaining greater understanding of their bioavailability, pharmaco logically effective concentrations and inter-individual variations in fish. We quantified plasma prostaglandin (PG) and plasma NSAID concentrations in individual female rainbow trout during and after 12 days continuous flow-through exposures to ibuprofen (0, 10, 200 mg/L) or diclofenac (0, 5, 100 mg/L). High-level NSAID exposures significantly reduced plasma PGE2 concentrations, but not other testable factors. The anticipated inter-individual variation of PGE2, 2.6-1.43 pM for ibuprofen; 0.8-1.88 pM for diclofenac; versus 0.8-3.16 pM in control fish. There was no significant correlation between plasma PG and plasma NSAID concentrations within exposure treatments; plasma NSAID concentrations exhibited much lower inter-individual variation, with blood plasma: water partition coefficients ranging from 1-3 for ibuprofen and 1-9 for diclofenac. To identify factors affecting PG levels in individual fish we measured plasma lipid content and plasma protein binding influencing partitioning and bioavailability, haematocrit and reactive protein concentration quantifying baseline immune system status, and plasma cortisol concentrations as a measure of stress in fish, potentially affecting plasma NSAID and PG concentrations. From our analyses, no single factor could explain the observed variations in NSAID uptake and pharmacological response. Our data highlight some of the complexities in interpreting biological exposure and effects data for NSAIDs.

455 Environmental effect assessment of human pharmaceuticals - the regulatory way forward

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Human pharmaceuticals are extensively studied and assessed before marketing approval. The EMA guideline for environmental risk assessment of human pharmaceuticals (EMEA/CHMP/SWP/4547/02) was adopted in 2006 and is currently under revision. Input has been provided by several stakeholders from academia, industry and government. In this context, the UBA experiences with effect based assessment of human pharmaceuticals will be evaluated and presented. The basis for the evaluation are double quality checked effect data of algae, aquatic invertebrates and fish provided within several European authorization procedures. Based on biodiversity, interest of the examined species and data sets, a prioritisation procedure was anonymous and encoded form. One discussion point will be the question whether the current base of data is sufficient to draw general conclusions. Although the results are based on more than 10 years of experience with environmental risk assessment within the authorization of new human medicinal products, the data basis is still lower than desired. So for some pharmaceutical ingredients detected in surface waters environmental effect data are lacking, because they entered the market before implementation of the EMA guideline. Furthermore, the tailored assessment approach for substances with very specific mode of action will be addressed, especially regarding the remaining uncertainties for protection of biodiversity and the environment. This does apply e.g. for endocrine active substances like contraceptive or anti-cancer drugs. Furthermore, the evaluated data allow discussion about effect sensitivity of several taxonomic groups, as well as about the PEC action limit (10 mg/L) for effect data justification. The quotient between the effect values of most sensitive and most insensitive species in more than 20% of the evaluated cases is greater than 100. Fish are the most sensitive organism group in more than half of the cases. In this context, the proposed application of the acute no-effect concentration (AEC) to replace long-term toxicity data with a single compound was rejected since only the AEC is applied usually for chemicals without any specific mode of action will be analyzed.
ciprofloxacin, ampicillin, clocicloxacin, sulfamethoxazole, trimethoprim and pseudoepephrine) and 4 PCR ingredients (sodium lauryl ether sulphate, alcohol ethoxylates, ammonium thioglycolate and dichlorvos). This is the first attempt to prioritize PPCPs in Nigeria and it provides a useful priority set of chemicals for source water monitoring in the region. Future work will focus on evaluating the results of the prioritisation approach against real world monitoring data for Nigeria.

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Aquatic toxicity related to pharmacological or secondary targets of human pharmaceuticals
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Human pharmaceuticals target specific biological structures to exhibit their intended therapeutic effect. The presence of the anticipated biological target of a pharmaceutical in a non-target species may lead to specific effects in that organism, while in the absence of the target non-specific baseline toxicity such as narcosis would prevail. Yet, pharmaceuticals often do not only interact with the anticipated pharmacological target in patients, but can also interact with secondary targets. Hence, specific toxicity could occur in non-target species also in the absence of a conserved pharmacological target simply because the secondary target is conserved in that species. The present study explored this hypothesis testing anti-histamines as model substances in Daphnia magna and the green alga Raphidocelis subcapitata. Acknowledgment - The research leading to these results has received support from the Institute for Environment Research, RWTH Aachen / Department of Ecosystem Analysis ESA; R. Hamann, Fraunhofer IHE; S. Wuester, Institute for Environmental Research RWTH Aachen; M. Krauss, Helmholtz centre for environmental research - UFZ - Effect-Directed Analysis; M. Fenske, Fraunhofer Gesellschaft / Translational Medicine and Pharmacology; I. Werner, Ecotox Centre Eawg-EFPL / Department of Anatomy Physiology and Cell Biology; H. Hollert, RWTH Aachen / Institute for Environment Analysis ESA; H. Hollert, RWTH Aachen / Institute for Environment Analysis ESA; H. Hollert, RWTH Aachen / Institute for Environment Analysis ESA; H. Hollert, RWTH Aachen / Institute for Environment Analysis ESA.

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Neurotoxicity testing approach to investigate venlafaxine and oxazepam modulation of transcriptomics and behavioral profiles in zebrafish embryos and larvae
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Pharmaceuticals enter the aquatic environment largely through patient use, and result in a pseudo-persistent background in the aquatic environment. The risk of these compounds is assessed for new products registered since 2007. However, there is a legacy of essential medicines for which we need to understand more. Since most of the thousand or so pharmaceuticals used by patients are likely to pose little environmental risk, it is important to identify those of most concern in order to prioritise effort and resources; it is vital to be able to predict internal concentrations in aquatic organisms. One method to assess uptake potential is to expose the animals of most concern. However, there are potentially thousands of compounds to be prioritised. Factoring that we already know the uptake rate can be influenced by the concentration of the compound in the water, there are enormous ethical implications for conducting this work with live animals (fish), and significant cost of resources to practically conduct the work. Alternatives are clearly required. Significant efforts to better predict environmental exposure are underway as part of the iPiE project (IM grant no. 115735—iPiE). Those in silico methods provide a first tier of screening, but we are likely still faced with hundreds of compounds to assess at multiple concentrations. We have been developing in vitro tissue micro-organs (organoids) that replicate the in vivo tissue. These can be used to build a virtual fish that will allow the screening of pharmaceuticals (or other compounds of concern or even metals) without testing live fish (BBSCR/NERC grant BB/L01016X/1). By building fish tissue cultures that better represent the complexity of the in vivo situation, we are able to offer in vitro models that can simulate live fish. Water exposure to the gill model can now be tolerated (without compromise) for several weeks, intestinal models are similarly robust. Since both methods employ a permeable barrier culture, rates of flux can be measured that provide not only simple information such as a compound may be taken up, but also rates of uptake and excretion. These data can be used to build kinetic models. The liver spheroids provide a metabolic tissue that when used in co-culture with the gill or gut provide a simple virtual fish alternative to live fish. These methods offer a critical step between predicting compounds of highest concern and prioritising which require further testing.

Emergence and multidimensional interactions of engineered nanoparticles in toxicity

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Effects of fullerene C60 increasing concentrations in Mytilus galloprovincialis: role of mTOR in cellular/tissue alterations
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Little is known about the effects at cellular, tissue and individual levels of emerging contaminants such as fullerenes. In particular, the mechanisms of action of C60 are poorly investigated. In this research, the effects of C60 on mTOR (mechanistic Target of Rapamycin) activity in mussel digestive gland were studied. mTOR is an evolutionarily-conserved serine/threonine protein kinase that senses and integrates a variety of cellular physiological and environmental signals to regulate cell growth. mTOR is found in two functionally distinct complexes, mTORC1 and mTORC2. In particular, the phosphorylated active form of mTORC1 mediates temporal control of cell growth by activating anabolic processes (such as transcription, ribosome biogenesis, protein synthesis), and by inhibiting catabolic processes (such as autophagy); mTORC2 is primarily involved in actin cytoskeleton reorganization. Mussels were exposed to C60 (0.01, 0.1 and 1 mg/L) for 72h. Tissue C60 accumulation was evaluated by immunofluorescence using a specific antibody as well as by chemical analysis. Immunohistochemical analysis revealed the presence and cellular distribution of C60 in mussel tissues, already at the lowest concentration. Our data demonstrated that the changes of the phosphorylation of mTORC1 and mTORC2 may explain most of C60 effects studied at cellular and tissue level. Indeed, the C60 induced dephosphorylation of mTORC1 contributed to increase autophagy and to decrease protein synthesis as mTORC2; in particular, the reduction of lysosomal/cytoplasmic volume ratio of the digestive gland cells; and mTORC2 to affect cytoskeleton organisation as revealed by the changes of actin/tubulin structures. Transcriptomic data are important to understand the cellular adaptive responses to the chemical. For this purpose, a novel low density oligo microarray (470 genes, suitable to follow 15 stress response pathways) was used. Transcriptomic analysis identified a number of DEGs showing a bell-shaped trend with a maximum in animals exposed to 0.1 mg/L C60. In terms of processes related to the DEGs depicted in all conditions, the most affected are associated to

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Virtual fish tales: Liver, Intestinal and Gill Organoids as an in vitro alternative to live fish for prioritising pharmaceuticals and other compounds of highest concern in the environment.

Transcriptomic analysis identified a number of DEGs showing a bell-shaped trend with a maximum in animals exposed to 0.1 mg/L C60. In terms of processes related to the DEGs depicted in all conditions, the most affected are associated to

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translational, cytoskeleton organization and mitochondrial activity. Transcription of selected genes was verified by RT-qPCR. These represent the first data on C60 tissue subcellular distribution and on the possible involvement of mTOR in the physiological alterations due to nanoparticle accumulation.

461 Protonic responses to nanoparticleulate and ionic silver in freshwater microbes with different background D. Barros, Universidade do Minho / Centre of Molecular and Environmental Biology, Department of Biology; A. Pradhan, University of Minho / Department of Biology; P.M. Santos, C. Pascoal, F. Cassio, University of Minho / Centre of Molecular and Environmental Biology CBMA Department of Biology. Enhanced use of AgNPs (AgNPs) has inevitably resulted in their release into freshwaters raising concern about the risk to non-target biota and related ecological functions. Functional protonics is an emerging technology that provides high-throughput analyses augmenting measurements of direct and highly sensitive responses at the cellular and sub-cellular levels. The impacts of AgNPs and ionic Ag at EC20 (effective concentration) were assessed based on the variations in the overall proteome in two aquatic fungal strains of *Articulospora tetracaulida*, one isolated from a non-polluted stream (At72) and the other from a metal-polluted stream (At61), and the bacterial strain *Pseudomonas* sp. M1 (PsM1) isolated from a metal-polluted stream. At72 was the most sensitive to AgNPs, whereas PsM1 was the most tolerant one. Characterization of AgNPs showed increased particle stability and lesser agglomeration with time at At72 while for At61 and PsM1 there was an increase in AgNPs agglomeration explaining its lower impacts on their growth. In fungi, 40% of the total quantified proteins were significantly altered after exposure to AgNPs and Ag2+ whereas for PsM1 this percentage was lower (~20%). At72 and At61 shared only 20% of the proteins suggesting that the biological pathways involved in Ag+ and AgNPs exposure were different. At61 had ~25% more proteins induced by both Ag forms (compared to At72), suggesting higher response which is consistent with the background of this fungal strain. In PsM1, 32% of the proteins increased under exposure to AgNPs whereas the percentage for Ag+ was higher (68%) indicating different responses to Ag+ and AgNPs. In At72, Ag+ increased the content of proteins involved in protein homeostasis while AgNPs increased the content of proteins related to DNA repair, the transport of substances and energy production. In At61, AgNPs increased the content of proteins involved in protein synthesis and energy production while both forms of Ag increased the content of proteins related to cell-redox and protein homeostasis, biomass and spores production and also to nucleic acids metabolism. Both Ag forms induced stress-responsive proteins which was consistent with the responses of enzymes involved in oxidative stress. Overall, functional protonics can be useful to get a mechanistic insight on the stress induced by AgNPs and Ag+ in microbes that play key roles in freshwater ecosystems.

462 Hazard assessment of seven different commercial silica nanoparticles on a battery of test species: bacteria, algae and fish cell lines P. Kulok, University of Göttingen / Department of Biological and Environmental Sciences; J. Sturve, Goteborg University / Department of Biological and Environmental sciences; T. Backhaus, University of Gothenburg / Department of Biological and Environmental Sciences. Manufactured silica nanomaterials are widely used in numerous applications in society such as paints, coatings, cosmetics, textiles and food and its release into the environment is of concern. This has raised the concern regarding the risk of silica nanomaterials and was in 2010 selected as one of the priority substances by the OECD Working Party on Manufactured Nanomaterials. In order to thoroughly examine the toxicity of silica nanomaterials to deuteriums, primary producers and fish, a panel of seven well characterized (with different size, coating and charge), biocide free, silica nanomaterials, were tested on bacteria, algae and fish cell lines. Based on the result, the current study also examined the selection of an appropriate exposure metric comparing mass (mg/L), number of particles (No/L) and surface area (m^2/L) against the observed toxicity. The results show that gill cell lines were the most sensitive test model with the lowest reported EC20 value of 5.1mg/L after exposure to the smallest particle at a concentration range of 12.5-100mg/L. Toxicity to fish cells was determined to be surface dependent, except for particles coated with ethoxy silane, which did not show toxicity. For bacteria and algae, the cell wall seems to play a major role in the uptake and toxicity of silica nanoparticles. Keywords: hazard assessment, silica nanoparticles.

463 Toxicity Assessment of Engineered Titanium Dioxide Nanoparticles S. Bitragunta, Birla Institute of Technology & Science Pilani, Hyderabad Campus / Biological Sciences; S. Palani, Birla Institute of Technology & Science, Hyderabad Campus / Biological Sciences. Titanium dioxide engineered nanoparticles (TiO2-ENP) are extensively employed in manufacturing of cosmeticceuticals, pharmaceuticals and health care products. As a result, TiO2-ENP can reach the ultimate sink such as soil in the environment during their life cycle. In this context, investigations to understand environmental implications of nanoparticles including TiO2-ENP are gaining prominence across the globe. In the backdrop of assessment toxicity of rutile TiO2-ENP (r-TiO2-ENP) in soil sentinel, present study is aimed at evaluating their toxicity as per OECD-207 guidelines on earthworm, Eisenia fetida. Physicochemical characterization of r-TiO2-ENP using dynamic light scattering revealed their tendency to form agglomerates (330-480 n.m) in water. Soil exposure of earthworms to r-TiO2-ENP (0.1, 0.15, 0.2 and 0.25 mg/kg) showed no mortality after 48 h. Increased specific activities of antioxidant enzymes including catalase, superoxide dismutase and glutathione peroxidase as well as lipid peroxidation indicate the potential of r-TiO2-ENP to induce oxidative stress in the sentinel organism. Interpretations of the study can serve as cues to design a comprehensive approach for developing invertebrate based biomarkers and indicators as early warnings for assessing environment and health impacts of engineered nanoparticles.

464 Combination effects of chlorpyrifos and ZnO on oxidative stress and reproduction of the earthworm Dendrobaena veneta D. Hackenberg, Department f Biology, University of Osijek / Department of Biology; L. Antic, University of Osijek / Department of Biology; D. Markovic, University of Rijeka / Department of Biotechnology, 1. ObboDzaz, Rudjer Boskovic Institute; B. Hackenberg, Department f Biology, University of Osijek / Department of Biology. When assessing the toxicity of pesticides and other chemicals to non-target organisms the most common experimental set-up is an exposure to a single compound. However, it is more likely for organisms under the environmental conditions to be exposed to a combination of different chemicals that have different modes of action and potentially can interact with each other. The aim of this research was to investigate the effects of a mixture of ZnO and chlorpyrifos (CHP) on biochemical biomarkers and reproductive success of the earthworm Dendrobaena veneta with possible differences in effects of ZnO in nano and bulk form, and the difference of effects in various soils. EC50 values for reproduction of D. veneta after exposure to CHP and ZnO were calculated and used in the binary toxicity experiment. Concentrations were as follows, with EC50 being 100%: 100% CHP; 75% CHP/25% ZnO, 50% CHP/50% ZnO, 25% CHP/75% ZnO, 100% ZnO. ZnO was separately tested as bulk and nano sized particles and CHP was tested as a commercial preparations. The entire experiment was conducted according to the OECD earthworm reproduction test. At the end of the experiment the number of juveniles and activities of AChE, CAT, GST, TBARS, MT and PC were measured. The concentrations of pesticides in soil samples were measured with UPLC and the concentrations of Zn was measured with the energy dispersive x-ray fluorescence technique (EDXRF). CHP had a clear effect on AChE activity with almost 50% inhibition after 28 days of exposure in artificial soil. CAT activity did not change on any concentration, for both ZnO and ZnO2, while GST activity decreased. The number of juveniles was significantly reduced with all mixture ratios. In natural soil CAT activity was also unaffected with bZn, and slightly induced with ZnO/CHP combination. The inhibition of AChE was present at all applied combinations. In natural soil the number of juveniles was reduced with ZnO, while it remained the same as control after bZnO exposure. The effects of combined exposure of CHP and ZnO on Dendrobaena veneta must be used wisely if the development of the sector is to be sustainable. Appropriate environmental characteristics, good water quality, well-understood social interactions and use of inland and coastal resources are essential to maintaining in existing and future aquaculture systems. Consequently, suitable zoning, selection of sites and application of carrying capacity are among the most important issues for the future success of European aquaculture, and also predicate the need for sustainability, resilience and best practice guidelines, as provided by the Ecosystem Approach to Aquaculture. The four-year Horizon 2020 TAPAS research project, which started in March 2016, aims to consolidate the environmental sustainability of European
aquaculture by developing tools, approaches and frameworks to support EU Member States in establishing a coherent and efficient regulatory framework, implementing the Strategic Guidelines for the sustainable development of European aquaculture and delivering a technology and decision framework for sustainable growth. The ultimate goal of the project is to create cost-efficient management tools and practices for the European aquaculture sector to investigate the scope of fish farming activity, social interactions, potential environmental impacts and any future risks.

467 Preliminary investigation on the occurrence of multifunctional organic micropollutants in offshore seawater and fish farm waters in Singapore, Vietnam, China, National University of Singapore / Civil & Environmental Engineering

Limited research has been conducted on the occurrence and distribution of antibiotics, pharmaceuticals, personal care products, endocrine disrupting chemicals and artificial sweeteners in the marine environment despite being increasingly impacted by these micropollutants (MPs). In this study, the presence and distribution patterns of multifunctional organic micropollutants in different fish farms and seawater samples were investigated. The sampling area is affected by various anthropogenic pressures including treated effluents, fish farming, shipping and port activities. A total of 23 MPs were found in offshore seawaters, 9 of them with detection frequencies higher than 50%. The highest detected values corresponded to cyclamates, salicylic acid and sucralose, with concentration range of

468 Perspectives on Urbanization, Water Reuse, and Aquaculture Product Quality

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By 2050, it is estimated that global food production must increase by 50%. Aquaculture will play an important role in meeting these needs. For example, in 2014 aquaculture surpassed global fisheries in providing food for human consumption. It is important to note that global aquaculture activities can in urban and periurban regions with differential waste management capacity. Yet high population densities in urbanizing regions result in concentration of food, energy, water and other resource consumption. Urbanization also leads to concentration of chemical use, which inherently results in exposures to human populations and ecosystems receiving waste streams within and from these urban centers. In developing nations, where many of the megacities will continue to emerge over the next few decades, access to chemical products is occurring faster than public health interventions and environmental management systems are being implemented. Unfortunately, 80% of the global sewage production is not treated, but returned to the environment and thus reused for various purposes. These non-traditional reused waters are being recycled for agriculture, including aquaculture in areas experiencing rapid urbanization, and then implications for water security, food safety and international trade are not routinely examined to manage more sustainable aquaculture practices. In the current presentation we draw from our ongoing efforts in Asia and North America to understand bioaccumulation of organic contaminants of emerging concern in common fish and shellfish used for aquaculture. For example, we have observed aquacultured bivalves to accumulate diverse contaminants of concern (e.g., pharmaceuticals, pesticides, flame retardants), apparently from landfill leachates and other diffuse sources such as sewage inputs. Furthermore, we have examined the interactions among water reuse practices and aquaculture for various products. Such efforts appear warranted at the global scale.

469 Bioaccumulation of selected veterinary medicines in the blue mussel (Mytilus edulis)

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Veterinary medicines are widely used within the fish farming industry for the control of sea lice infestation. In 2016, over 10 tonnes of veterinary medicines were used by Norwegian fish farms for the control of sea lice. The impact on non-target species has raised increasing concern. For instance, do wild and farmed mussels in the vicinity to these fish farms have the potential to bioaccumulate these chemicals and thereby pose a threat to human health? On the other hand, mussels may be the most suitable biomonitoring species for the presence of veterinary medicines in the environment. To better understand these scenarios, and the fate of these chemicals in the environment, a series of laboratory controlled exposures were performed to determine the bioaccumulation and depuration of selected veterinary medicines in the blue mussel (Mytilus edulis). The veterinary medicines included teflubenzuron, emamectin, deltamethrin and azamethiphos. Due to the low solubility of teflubenzuron and deltamethrin a saturation column was employed within a flow-through system to deliver a stable concentration of test chemical over a 14-day uptake phase. Water and mussel samples were collected at time intervals during the 14-day uptake phase, and again following transfer of the mussels into clean flowing seawater during the 7 to 14 day depuration phase. The effects of salinity on the bioaccumulation and depuration of teflubenzuron also differed in that other mussels in brackish waters show different bioaccumulation dynamics. So far, results have shown a clear uptake of teflubenzuron over 14 days, reaching maximum concentrations (~1500 ng/g) after 10 days. Depuration of teflubenzuron was fast for the first 2 days, although still present at approximately 250 ng/g after 7 days depuration. Salinity had no apparent effect on the bioaccumulation of teflubenzuron. In contrast, deltamethrin showed lower bioaccumulation, with maximum concentrations of 45 ng/g after 6 days. No significant depuration of emamectin was observed after 7 days in clean flowing seawater. The results suggest that mussels are a suitable biomonitoring species for the presence of veterinary medicines in the environment. Additionally, mussel farms in close proximity to fish farms have the potential to bioaccumulate these chemicals in their tissues and is subsequently recommended for monitoring.

470 Contribution of nuclear applications to better understand bioaccumulation of contaminants in aquaculture species

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Environmental pollution from aquaculture is often seen as a major concern, but today, increasingly is the potential exposure of aquaculture to contaminants. In order to fully understand the contamination risk of farm-raised species, nuclear applications can be used. This is a very powerful approach that allows identifying the susceptibility of economically important species (fish and seafood) to be contaminated. Marine fish farming is regarded as the future of aquaculture and thus, the safety of these farm-raised fish is paramount particularly as 50% of fish consumed are now farm-raised. Therefore, a better understanding bioaccumulation processes of such contaminants with current aquaculture practices is essential. Such work will attempt to better understand the role the fish food or key environmental parameters on contamination of fish that may affect the health of the farmed species and/or the human consumer. This has been commonly done in a natural setting but is now beginning to be examined for fish farming practices. Major advantages of radionuclides over conventional techniques are their very high sensitivity and discrimination capacity; it permits the measurement of bioaccumulation kinetics of several elements at realistic (viz. low) environmental concentrations in a single experiment. Furthermore, some radionuclides permit the non-destructive analyses of contaminant levels in living organisms. This paper identifies present and future threats on farm-raised fish from a contamination point of view, and presents a synthesis of experimental results completed on farm-raised fish using radionuclides to contaminated environments. For example, the various effects that food, water salinity and temperature can have on the assimilation efficiencies of trace elements and radionuclides in farmed fish. Such findings paving the way for further investigations on the potential use of nuclear techniques in aquaculture and food safety.

471 Effects of antibiotic’s medicated fish feed in the marine environment

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Intensive aquaculture is considered to be an important source of antibiotics into the marine environment. Antibiotics used in aquaculture have been reported to accumulate on sediments and non target aquatic organisms, modifying the biodiversity and the environmental conditions in areas close to fish farms. Moreover, recent studies show that aquaculture may contribute to the emergence of resistance genes in environmental bacteria next to fish farms, which indicates the assembly, selection and dissemination of antimicrobial resistance through open aquaculture installations and might thereby compromise environmental and human health. A field experiment was performed in a moderately impacted bay in the south east coast of Spain (Aguilas, Murcia), which consisted of a series of sediment traps (concrete) covered with local species of mussel (mytilus edulis) medicated or non medicated with three antibiotics (oxytetracycline, florfenicol and flumequine). Fish eggs were applied simulating fish farm losses for a period of 3 weeks. Measured antibiotic concentrations in the sediment were 2700 – 8000 ng/g (average 1% of the applied amount) for oxytetracycline, and 19000 – 54000 ng/g (average 10% of applied amount) for flumequine. Florfenicol was not detected. Different accumulation rates were found in covered/uncovered traps due to wild fish influences in the availability of feed and bioturbation. Physico-chemical characteristics of the sediment also changed; with a higher S and lower N content and a larger percentage of fine material in feed affected treatments. Invertebrate
presence was also correlated with the food availability, although no evident effects of the antibiotics were found over the analyzed samples. Bioaccumulation of the target antibiotics in the invertebrate community and evaluation of the antibiotic impacts over the microbiome and resistome of the sediment bacteria is still ongoing. This is one of the first studies describing fish feed and antibiotic impacts produced by aquaculture under Mediterranean conditions.

Systems ecotoxicology: application of OMICS data across multiple level of biological organization in research and risk assessment (II)

472 Systems toxicology approach for the assessment of zebrafish cardiac and neurotoxicity

A major goal in the field of toxicology is to predict long term animal health risks and/or environmental hazards associated with a particular substance(s).

Traditionally utilised classical toxicology methods involve animal exposure over a relatively short period and recording adverse outcomes. These data are then extrapolated to humans and to other species. The accuracy of such extrapolation would benefit from mechanistic understanding of toxicity. However, molecular basis for adverse outcomes is not easily interpreted from classical toxicology methods. Here we present our systems toxicology approach that focuses on deciphering biological mechanisms responsible for adverse outcomes. The underlying structure of this approach is a computable biological network model. We have developed two models describing molecular pathways that lead to cardiotoxicity and neurotoxicity in zebrafish larvae based on the knowledge curated from scientific literature. Key signalling nodes in the model are linked to information about downstream gene expression. Differential expression of downstream genes can be used to infer activity of the upstream protein – a process termed network scoring. Scoring of the network highlights the most affected nodes, which leads to mechanistic hypothesis generation and gives a quantifiable measure of network perturbation. In parallel to network scoring, we utilize classical toxicology methods to detect adverse outcomes. We present the acute toxicity results for selected chemicals (e.g. acrylamide, arsenic, citolopram, imidacloprid) according to the OECD fish embryo acute toxicity test (OECD test guideline 236). We then report results from chemically exposed larvae in functional cardiac and behavioural assays, and transcriptomics analyses. Finally, we describe the utility of the network model in interpreting transcriptomics analyses to gain mechanistic insight into the molecular events initiated by a given chemical. Cardiac and neural apical endpoints together with computational network scoring provide a comprehensive method for linking molecular events to organ toxicity. This approach will enable more accurate toxicity predictions over long exposures and in different species.

473 Time response relationship between gene expression and life history in a Daphnia population exposed to heavy metals
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Over the last decade, molecular technologies have evolved into robust high throughput platforms available to many scientists in a wide variety of disciplines. Implementation of these technologies in ecotoxicology and risk assessments have focused on mechanisms of toxicity and stress response on the gene level to explain effects at the organism level. However, current studies remain focused at the individual level and rarely include population level molecular responses. Population level molecular responses may provide a better insight into the potential mechanisms at play at the population level while at the same time avoid focusing on gene expression patterns that are the cause of clonal or interindividual variation. Furthermore, most studies select an arbitrary timepoint and hence measure expression responses without any prior knowledge. Here, we focus on population level molecular responses of a Daphnia magna population to arsenic and copper and their binary mixture. The population was exposed to low chronic toxicity concentrations of arsenic and copper resulting primarily in effects on reproduction rather than survival. Rather than focusing on a single arbitrary timepoint, gene expression data and life history data were both recorded at multiple time points. As such, these datasets will provide a first basis on how exposure duration may affect the conclusions and decisions made about the toxicity of chemicals. In addition, by collecting both molecular data and life history data, we will be able to better understand the time response relationship in populations under stress both at the life history level and the molecular level. This will allow us to better integrate these two data types and identify potential causal relationships between the molecular level and the life history level. The identification of such causal relationships will play an integral part of incorporating omics data in environmental risk assessment.

474 How to implement functional responses of microalgae in risk assessment processing?
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Microalgae (e.g. bacteria, fungi and algae) are involved in various ecosystem functions such as biogeochemical cycles or pollutants degradation meaning that they are crucial for ecosystem functioning. In the environment, organisms are exposed to anthropogenic pressures which are known to potentially induce structural and functional changes. If such causal links are identified, little is known about the involved biochemical pathways supporting specific functions. Moreover, most of a priori ecological risk assessment (ERA) tools are based on structural endpoints and do not necessarily ensure the protection of these functions. The recent raise of OMICS approaches (e.g. transcriptomics and metabolomics) opens the perspective in ecotoxicology to explore pathways involved in ecological functions. The main aim of this study was to provide a new and innovative risk assessment tool based on functional responses (captured via OMICS approaches) of microalgae to chemical stressors.

The rationale to use OMICS in such context is to provide more protective and early warning thresholds. The transcriptomic and the metabolomic responses of Scenedesmus vacuolatus to triclosan were explored after exposure of 14 hours along an increasing gradient of 5 concentrations (from 0.69 to 6.63 µg/L, 5 replicates). Within a dedicated workflow, we selected the responsive molecular items through metabolites/transcripts that were concentration dependent on a number of them and we derived a sensitivity value from each curve (even the non-monotonic one). Molecular items showed mainly non-sigmoid and even non-monotonic responses to triclosan exposure. For example, the transcripts data were mainly best described by an exponential model for more than half of the curves and a Gaussian or log-Gaussian model for more than a quarter of the curves. Then, the molecular items were linked (when possible) to the pathways they are involved in. From that information, we built a SSD-like tool based on functional responses captured at the community level in order to protect functions and integrating two levels of OMICS responses. The next step consists to build such tool from the periphytic community level.

475 Sex, drugs and Daphnia magna. A multi-omics approach suggests conserved mechanisms of interaction between metallohormones and endocrine disruptors
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The assessment of environmental exposure to toxic chemicals released by human activity as well as their impact on biological systems is key to protect the biosphere. Currently, environmental risk assessment tools are based on chemical concentrations and do not take into account the assessment of biodiversity. Although this has been a very effective strategy, it has some shortcomings. These include the fact that a relatively limited number of compounds can be measured and linked to biologically relevant organism-level responses. The issue is particularly challenging in chronic exposures and in complex mixtures scenarios. This project aims at identifying the molecular networks linked to single and mixture exposures and to use these to infer the effects of chemical mixtures. We approached this important challenge by applying a systems biology approach to integrate expression profiling, metabolomics and phenotypic data (respiration and feeding rates), representing the response of Daphnia magna to a panel of environmentally relevant chemicals and their mixtures. Firstly, it was exposed to a mixture of single compounds with known mode of action (MoA) i.e. estrogen disruptors and acetylcholine esterase (AChE) inhibitors as well as metals with unclassified MoA. We have been able to model the differences between the two main MoA studied and linked them to biological activities within Daphnia. Furthermore, we have found that as expected, metals do not show a common MoA, with some of them clustering closer to either endocrine disruptors or AChE inhibitors (AChEi). Furthermore, the estrogen disruptors, has already been shown to play a role with the estrogen receptor in humans but its role in Daphnia is still under surveillance. To further study this finding we exposed D. magna to complex mixtures of Cd and ethynlesradiol. While the individual exposures triggered the alteration of expression of a relatively large number of genes, the exposure to the mixture showed little or no effect. These results indicated that both compounds share a complex interaction at a molecular level suggesting that the degree of conservation of the regulatory pathways underlying response to endocrine disruptors may be higher than previously thought. Overall, our work shows that it is possible to predict a compound MoA from its
molecular state and also predict additive or synergistic effects of mixture exposure.

476 Data-driven systems biology approach gives insight into a complex process of water remediation
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Introduction. Increasing population and industrial production put strain on clean water resources. Even in highly developed countries with advanced waste water treatment plants, water quality could be improved further before releasing it. Constructed wetlands have been used for water treatment for decades and are a low-cost natural option. Waterharmonica is an example of such additional treatment, consisting of a sedimentation pond, reed bed and a wetland forest. In this work, we have used three-spined stickleback living in mesocosms containing water from various stages of additional remediation from three different sites in the Netherlands. Aim. The aim of this work was to understand the effects of additional steps of water remediation. Results. We have used a data-driven systems biology approach to understand the relationship between the environment (chemical concentrations), molecular high-throughput measurements (stickleback liver gene expression), physiological parameters and more traditional measures of toxicity. We have integrated all different measurements into static similarity networks and modularised these so that in each module, genes are responding in a similar way during different stages of remediation. We see that some chemicals with high chemical risk (aldicarb, chlorpyrifos, fluoranthene, pirimiphos methyl) decrease in all sites and are also correlated with gene expression in both male and female stickleback. However, some chemicals are only correlated with gene expression in one sex or male of female stickleback. We also see that some chemicals for which predicted no-effect concentration (PNEC) is not known (such as PCB-56 and mineral oils) are associated with modules containing several high-risk chemicals. Functional annotation reveals further insights. For example a module of the male stickleback network correlated with liver weight and several chemicals including tricosan and phthalates has a statistically significant number of genes from the KEGG pathway „metabolism of xenobiotics by cytochrome P450“ but a module in the male-specific network that is not correlated with any chemicals, is enriched in KEGG functions related to immune functions ( „antigen processing and presentation” „type I diabetes mellitus“ and „cytokine-cytokine receptor interaction“). Conclusion. We have showed the potential of data-driven systems biology approach integrating various types of data to give insight into the effects of a complex remediation system.

477 Co-expression network analysis of massive proteogenomic data: applications in ecotoxicology.
D. Deligiannis, R. Arnaud, J. Arnaud / IRRELLERY Labortatoire Ecotoxicologie; C. ALMUNIA, CEAR París-Saclay; D.D. Gouveia, Irsée Lyon / UR MALY Laboratoire Ecotoxicologie; J. Trapp, Irsée Lyon; J. Gaillard, CEA / Laboratoire de Biochimie des Systemes Perturbés; O. Pible, CEA; a. chaumot, O. Geffard, Irsée / UR MALY Laboratoire Ecotoxicologie; J. Armengaud, CEA / Laboratory Innovative technologies for Detection and Diagnostics Dynamic and biocompatible microfluidic chips in test species under contaminant exposure promises the possibility to gain insights into the mode of action of chemical compounds and molecular pathways involved in toxic responses. Intuitive network concepts (e.g. connectivity and modularity) have been found useful for analyzing complex interactions and successfully applied to study gene-gene and protein-protein interactions. Currently, a majority of protein networks are constructed using protein-protein interaction (PPI) databases. However, manually curated PPI databases are typically heterogeneous, documented for few model species, and often characterized by incomplete coverage, and selection or detection biases. De novo (or no a priori) approaches based on observed data offer an alternative under which prior knowledge of protein interaction is not necessary but rather advantageously replaced by direct measurements and pair-wise correlation analysis of their abundance. This approach may be particularly powerful to identify signaling pathways which proteins with unknown function belong to or to identify novel, pertinent biomarkers of toxicant exposure. Here we present a network analysis method applied to shotgun high-throughput proteomic data we produced for the aquatic sentinel organism Gasterosteus aculeatus. Shotgun proteomics was used to identify the molecular key players involved in different physiological states linked to reproduction and in case of exposure to insecticides potentially inducing endocrine disruption in this crustacean. We identified protein reproduction and in case of exposure to insecticides potentially inducing endocrine molecular key players involved in different physiological states linked to exposure. Here we present a network analysis method applied to shotgun unknown may be particularly powerful to identify signaling pathways which proteins with interaction is not necessary but rather advantageously replaced by direct networks are constructed using protein data mining of high throughput omics data acquired in test species.

478 How researchers can work in alliance with citizens to fight misinformation and improve public debates
S. Vanhoenacker, Sense About Science EU
Public resistance against glyphosate, GMOs, animal testing, vaccination and numerous other scientific innovations has made many scientists defensive and paranoid about the public. Recent discussion about a post-truth society and anti-intellectualism have increased this perception of a hostile and ignorant public. With concrete examples, Sofie will illustrate a different, more effective approach for both researchers and non-researchers to bring back reason into emotional debates. This approach, called public expert-fed – in which scientists respond directly to real, unedited questions from the public – breaks through polarised and difficult debates because conversation is led by the questions and issues people raise. It allows researchers to identify gaps and misunderstandings in the public debate and to respond to them. With this approach, rather than fighting the public resistance against new technologies, researchers can work in an alliance with citizens to fight misinformation and improve the public debates.

479 Discussion: the need to promote good science and evidence in public debates

480 How to communicate the risks posed by endocrine disrupting chemicals? (I)
J. Legler, Utrecht University / Institute for Environmental Studies
There is emerging evidence that some of the increasing occurrences of endocrine-related disorders in humans and wildlife are linked to the exposure to endocrine disrupting chemicals (EDCs). Because of the potential of significant impacts on organismal and population health that can result from disruption of hormone homeostasis, numerous governments have established legislations that regulate chemicals that have the potential to interact with the endocrine system of humans and wildlife. However, while there is agreement on the need for regulation of EDCs, the frameworks to assess and regulate candidate EDCs differ significantly among regions. Furthermore, the mixed messages delivered by the media to the public with regard to the risks EDCs may pose add to the confusion currently existing within society, and which has split opinions on how to address this issue. This presentation will review the issue of endocrine disruption from a human health and environmental perspective, and discuss current approaches to the assessment of the risk/hazard of EDCs in Europe and North America. Through discussions among the presenters and the audience we aim to explore a roadmap on how to address the risks posed by EDCs and where the priorities for future research should lie.

481 How to communicate the risks posed by endocrine disrupting chemicals? (II)
M. Hecker, University of Saskatchewan / School of the Environment & Sustainability and Toxicology Centre
There is emerging evidence that some of the increasing occurrences of endocrine-related disorders in humans and wildlife are linked to the exposure to endocrine disrupting chemicals (EDCs). Because of the potential of significant impacts on organismal and population health that can result from disruption of hormone homeostasis, numerous governments have established legislations that regulate chemicals that have the potential to interact with the endocrine system of humans and wildlife. However, while there is agreement on the need for regulation of EDCs, the frameworks to assess and regulate candidate EDCs differ significantly among regions. Furthermore, the mixed messages delivered by the media to the public with regard to the risks EDCs may pose add to the confusion currently existing within society, and which has split opinions on how to address this issue. This presentation will review the issue of endocrine disruption from a human health and environmental perspective, and discuss current approaches to the assessment of the risk/hazard of EDCs in Europe and North America. Through discussions among the presenters and the audience we aim to explore a roadmap on how to address the risks posed by EDCs and where the priorities for future research should lie.

482 Discussion Endocrine Disrupting Chemicals

483 A regulator’s perspective in involving stakeholders and the public in the regulation of a substance
C. Ajao, ECHA-European Chemicals Agency; W. de Wolf, ECHA / Product Safety & Regulatory Compliance
The European Chemicals Agency (ECHA) was established in June 2007 through
the REACH Regulation, and the registration of all substances already on the market above 1 ton per annum will be completed in 2018. Since its start the areas of responsibility expanded from industrial chemicals to biocides, capturing as well the communication of chemical hazards to workers and the public through the Classification, Labelling and Packaging Regulation (CLP), and the regulation of international trade of hazardous chemicals. The latter includes support for the protection of human health and the environment by providing developing countries with information on how to store, transport, use and dispose of hazardous chemicals safely through the Prior Informed Consent Regulation (PIC). 'In its decision making and opinion forming, ECHA uses the scientific information provided by academia and industry and applies them within the regulatory framework that it operates. Transparency is one of the values that is driving ECHA in its interactions with its different stakeholders, and the ECHA Scientific Committees invite ECHA’s accredited stakeholders as regular observers and contributors to its meetings. 'In this presentation will look at regulatory science communication by describing the different stakeholders that ECHA interacts with, the forms of communication used and their timeframes. It will also explain the regulatory boundaries ECHA has to abide to, which influence the uptake of the latest science developments, and their communication with a special focus on the decision making and opinion forming at the Member State Committee. DISCLAIMER: ‘The views expressed in this abstract are solely those of the authors and the content of the paper does not represent the views or position of the European Chemicals Agency.'

484 Questions/Discussion

485 General Discussion with panel of Sofie Vanthournout, Juliette Legler and Markus Hecker

486 Concluding remarks part I and a teaser for part II!

487 The impact of chemical pollution on the resilience of soils under multiple stressors

488 Ecological risks under complex, multiple-stressor threat scenarios: integrating chemical effects with environmental drivers (III)

489 The effect of water chemistry on cadmium induced olfactory impairment in juvenile rainbow trout (Oncorhynchus mykiss)

490 Combined effects of temperature and metal exposure on cell membrane fatty acid composition, lipid peroxidation, antioxidant capacities and desaturation and elongation transcriptome in freshwater fish

M. Fadlouli, INRS - Eau, Terre et Environnement / Centre Eau Terre Environnement; F. Pierron, Université de Bordeaux / UMR EPOC CNRS 5805; P. Couture, INRS / Centre Eau Terre Environnement

In this project, two freshwater fish commonly found in areas affected by metal contamination were acclimated to different temperatures (9 and 28°C for yellow perch (Perca flavescens) and 15, 25 and 30°C for fathead minnow (Pimephales promelas)) and exposed either to Cd or Ni during 8 weeks. At the end of exposures, we measured cell membrane phospholipid fatty acid composition, the activities of superoxide dismutase, catalase, glutathione-S-transferase, glutathione peroxidase (enzyme indicators of antioxidant capacities), the concentrations of glutathione (antioxidant) and malondialdehyde (indicator of lipid peroxidation (LPO)) as well as the transcription levels of desaturases (fads2, desg2, scd2) and elongases (elovl6, elovl5, elovl7). Both yellow perch and fathead minnow counteracted the effects of changes in acclimation temperature on cell membrane properties by remodelling their phospholipid fatty acid composition. Specifically, in the muscle of both species, polyunsaturated fatty acids increased in cold-acclimated fish compared to warm-acclimated fish, in agreement with the theory of homeoviscous adaptation. However, brain cell membrane composition was more conservative, especially in fathead minnows. Polyunsaturated fatty acids are more vulnerable to LPO than saturated fatty acids and metal contamination leads to oxidative stress. We therefore tested the hypothesis that temperature-induced changes in cell membrane polyunsaturation are accompanied by variations in LPO in metal-exposed fish. Unexpectedly, in both species, metal exposure itself affected membrane fatty acid composition. In yellow perch, the normal response of cell membrane composition to thermal acclimation was reversed by exposure to both metals. Yet, in spite of the high polyunsaturation level in warm-acclimated fish under Ni exposure, MDA concentration was the lowest, suggesting a massive response of the antioxidant system to fight against LPO. In fathead minnow, metal exposure also affected the membrane fatty acid composition of both tissues, but was more subtle than for yellow perch. We observed a mismatch between desaturation and elongase gene transcription and membrane composition. Overall, our results suggest that levels of control of cell membrane fatty acid composition other than gene transcription may be affected by temperature and metal exposure, such as post-transcriptional regulation of gene transcription and de novo phospholipid biosynthesis.
bioavailability. In conclusion, water chemistry is an important modulator of metal toxicity, not only for acute lethality but also for sub-lethal effects, such as olfactory impairment. In order to enable the prediction of site-specific olfactory toxicity, the development of a BLM parameterized to the olfactory system of fish would be very beneficial. However, more data on the effect of water chemistry on metal-induced olfactory impairment is required to be able to determine affinity constants and maximal binding capacities.

490 Physiological and biochemical responses of polychaetes: interplay of elements contaminated sediments and salinity changes
A. Pires, Universidade de Aveiro / Biologia; R. Freitas, University of Aveiro / Departamento de Biologia; CESAM; C. Patinha, Universidade de Aveiro, E.F.; Silva, University of Aveiro / Geosciences; E. Figueira, University of Aveiro / Biology CESAM

Coastal systems often serve as sinks for toxic elements, and changes in salinity, predicted to occur due to global climate change are believed to influence elements geochemistry in aquatic systems. The effects of these changes can also alter biota sensitivity and mediated effects led to a better bioavailability of trace elements in marine communities, mainly on those organisms living in sediments, such as polychaetes, which also support much of the diversity at higher trophic levels. So, this work examines the interactions of elements contamination and change in salinity and polychaetes performance, highlighting modifications that coastal systems may undergo due to alterations driven by salinity change. Polychaetes were exposed to elements contaminated sediments (median sand and fine sand sediments, collected from contaminated areas from ria de Aveiro lagoon, Portugal) and salinities 21, 28 and 40, for 28 days. This study aimed to evaluate physiological (regenerative capacity of Dicnatra nepolaolina), behaviour (burrowing tests with Hedite diversicolor and Arenicola marina) and biochemical responses: indicators of cell damage (LPO) antioxidant (SOD and CAT), and biotransformation (GSTs) enzymes. After exposure, both types of sediments had lower total elements concentration (TEC), when compared to original sediments, mainly in sediments with A. marina and H. diversicolor. Sediments exposed to salinity 40, mainly those containing H. diversicolor had even less TEC than remaining sediments, which seems that salinity changes may influence elements availability. LPO levels were higher at salinity 40 for H. diversicolor and A. marina, and at salinities 21 and 40 for D. nepolaolina. Although polychaetes were able to increase the activity of SOD, CAT and GSTs, these defense mechanisms were not sufficiently efficient to fight against the excess of ROS, leading to LPO. Furthermore, in H. diversicolor, the burrowing behaviour was impaired in polychaetes in fine sand sediments and salinity 40. For A. marina exposure to median sand sediment for all salinities and to fine sand for salinity 40 led to a decrease in bioturbation capacity in marine neopolaolina individuals exposed at salinities 21 and 40, for both sediments, exhibited lower capacity to regenerate their body when compared to salinity 28 (control). Overall, this study demonstrates that variations in salinity can strongly affect elements availability. Interaction of both variables impacted polychaetes responses differently.

491 Do trace metal contamination and parasitism infestation influence the activity of the bioturbator Upogebia pusilla?
A. da Ruirin, EPOC, University of Bordeaux / UMR EPOC CNRS 5805; X. de Montaudouin, A. Clurot, P. Gonzalez, University of Bordeaux / UMR EPOC CNRS 5805; A. Bagaud, Bandimont, Université de Bordeaux / UMR EPOC CNRS 5805; O. Maire, P. Goureus, G. Daffe, A. LEGEAY, University of Bordeaux / UMR EPOC CNRS 5805

In marine environment, bioturbating species are considered as important ecosystem engineer species. Bioturbators are mainly benthic organisms living in the sediment. Their fossorial life style deeply alters the physical and biochemical properties of sediments. In marine soft-bottom environments, mud shrimp are considered as among the most important bioturbators. Because of their intense burrowing activities, they exert a major influence on solute and porewater exchanges, habitat structuration and benthic community composition. The influence of mud shrimp on their environment is related to the intensity of their behavior. Several factors could interact with bioturbators fitness and therefore modify their influence as ecosystem engineer species. Regarding mud shrimp, only a few factors have been studied. Among them, the role of trace metals on mud shrimp fitness and bioturbation activities has never been investigated yet. Besides, mud shrimp are frequently engineer species. Regarding mud shrimp, only a few factors have been studied. Their fossorial life style deeply alters the physical and biochemical properties of sediments at salinity 40 led to a decrease of burrowing kinetics. Although polychaetes were able to increase the activity of SOD, CAT and GSTs, these defense mechanisms were not sufficiently efficient to fight against the excess of ROS, leading to LPO. Furthermore, in H. diversicolor, the burrowing behaviour was impaired in polychaetes in fine sand sediments and salinity 40. For A. marina exposure to median sand sediment for all salinities and to fine sand for salinity 40 led to a decrease in bioturbation capacity in marine neopolaolina individuals exposed at salinities 21 and 40, for both sediments, exhibited lower capacity to regenerate their body when compared to salinity 28 (control). Overall, this study demonstrates that variations in salinity can strongly affect elements availability. Interaction of both variables impacted polychaetes responses differently.

492 Integrating ecotoxicology and ecology to advance understanding and prediction in multiple stressor research
R. Schafer, University Koblenz Landau / Institute for Environmental Sciences; J. Pigott, Trinity College Dublin / Zoology

Global environmental change is driven by multiple anthropogenic stressors. Conservation and restoration requires understanding the individual and joint action of multiple stressors to evaluate, design and implement management strategies. To date, most studies on multiple stressor effects have sought to identify potential stressor interactions, defined as deviations from null models, and related meta-analyses have focused on quantifying the relative proportion of stressor interactions across studies. These studies have provided valuable insights about the complexity of multiple stressor effects, but remain largely devoid of a theoretical framework for prediction of effects and null model selection. We suggest that multiple stressor research would benefit by 1) integrating additional null models from ecotoxicology and 2) selecting null models based on their mechanistic assumptions of the stressor mode of action and organism sensitivities as well as stressor-effect relationships. We present a range of null models and outline their underlying assumptions and applications. Moving forward, this integration requires multiple stressor research to shift its focus from identifying statistically significant interactions to the use and development of mechanistic (null) models. We discuss how ecotoxicological and ecological concepts will aid in achieving this.

Improveing the Quality of Ecotoxicological Testing and Assessment

493 Updating the OECD Guidance Document 23 on aquatic toxicity testing of difficult substances and mixtures to include state-of-the-science approaches
W.S. Hunter, U.S. Food and Drug Administration / Center for Veterinary Medicine; G. Stoddart, C. Faltbender, PETA International Science Consortium Ltd.; M. Halder, European Commission Joint Research Centre / DG Joint Research Centre IHCP EURL ECVAM; E. Salinas, BASF SE / Eperimental Toxicology and Ecology

The Organisation for Economic Cooperation and Development (OECD) Guidance Document (GD) on Aquatic Toxicity Testing of Difficult Substances and Mixtures (GD 23) was first published in 2000 and provides crucial guidance that supplements OECD Test Guidelines. Since its release much experience has been gained in handling difficult-to-test chemicals in aquatic exposures as well as progress made in developing methods for testing difficult test chemicals. The GD was revised as recently as 2016 to include state-of-the-science approaches. We provide an overview of the updated GD 23. One significant revision was the expansion of the guidance on testing of poorly water soluble test chemicals. Attention was paid to updating exposure methods that do not employ a solvent in order to eliminate the need for a solvent control, and thus, reducing the number of animals used in aquatic toxicity tests. Another major revision was the addition of more detailed guidance for substances of unknown or variable composition, complex reaction products, and biological materials (UVCBs). The presentation also briefly describes other aspects of the updated GD of interest to those involved in aquatic toxicity testing. The updated GD 23 will help government agencies, industry, and contract research organisations conduct valid and reliable aquatic toxicity studies on difficult-to-test chemicals while minimising both the number of animals used and the need to repeat studies. The views, conclusions and recommendations presented in this presentation are those of the authors and do not necessarily represent the policies or positions of the United States Food and Drug Administration, the PETA International Science Consortium Ltd., the International Council on Animal Protection in OECD programmes, the European Commission or the OECD.

494 Calibrating Non-Target Arthropod (NTA) Lower Tier Assessment Factors
F.M. Bakker, Eurofins-Mitox; S. Aldershof, Bioresearch and Evaluation; A. Dinter, Cheminova Deutschland GmbH & Co. KG / Global Regulatory Sciences; E. Elston, Syngenta Ltd; C. Mayer, BASF SE / Ecotoxicology; E. Pilling, Dow Agrosciences / Regulatory Sciences; G. Weyman, ADAMA; P. Neumann, Bayer AG

Assessment factors for Tier 1 and Tier 2, with several test species and different test chemical structures, are used to determine the potential environmental risk of NTA. These assessment factors describe other aspects of the updated GD of interest to those involved in aquatic toxicity testing. The updated GD 23 will help government agencies, industry, and contract research organisations conduct valid and reliable aquatic toxicity studies on difficult-to-test chemicals while minimising both the number of animals used and the need to repeat studies. The views, conclusions and recommendations presented in this presentation are those of the authors and do not necessarily represent the policies or positions of the United States Food and Drug Administration, the PETA International Science Consortium Ltd., the International Council on Animal Protection in OECD programmes, the European Commission or the OECD.
outcome with other taxa were observed, these were considered jointly. As expected Tier 1 studies had the most sensitive endpoint and consequently the largest HQ. Using the recovery endpoint, it was found that for the off-field HQ’s of 1, 6 and 250 delimited recovery ranges of 0 weeks (no effects), 4 weeks and 12 weeks respectively in the off-field situation (hay meadow paradigm). For the in-field situation recovery intervals of 0-1, 1-2, 6-12 and 12 months were delimited by HQ-values of 40, 375, 620 and 2500. Tier 2 studies could have lethal or sublethal endpoints. Using the most sensitive of the two and including a Vegetation Distribution Factor (VDF) of 5 the following HQ-values were derived for the off-field: HQ=1.7 for a no effect level. These HQ’s also correspond to 1 and 2 month recovery periods. For the in-field, using VDF=1, HQ’s are 1.3 for no effect and 6.6, 15, 60 and 560 for 1, 2, 6 and 12 months respectively.

495 The unforeseen consequences for animal welfare of the OECD TG 240 (MEOGRT) biological validity criteria E. Salinas, BASF SE / Eperimental Toxicology and Ecology; L. Weltje, BASF SE / Crop Protection Ecotoxicology

The Medaka extended fish chronic reproduction Test (MEOGRT) was established in 2015 as OECD test guideline (TG) 240; a level 5 investigation under the OECD conceptual framework for endocrine disruption assessment. The MEOGRT brings together all aspects of the OECD chronic fish TGs 210, 234, and 229 into one test. OECD TGs include validity criteria as minimum standards for acceptable performance and particularly the biological control performance criteria are critical to assure relevant effects of a test chemical are detectable. Validity criteria assist regulators in determining study quality and reliability; studies that do not comply may be rejected and/or repeated. Compared to other fish TGs, the quantity of validity criteria in the MEOGRT TG have increased and are more stringent, thus elevating the potential for failure and repetition. Other investigators has already noted a high incidence of study repetition following well established OECD fish TGs. However, data as is available for the MEOGRT is currently very few laboratories can implement this highly complex TG. The MEOGRT arose from an international validation effort and recently the data from 9 validation studies were published. We compared control performance in those studies against the existing MEOGRT validity criteria to evaluate the compliance rate. Only 3 studies reported the control parameters corresponding to all biological control criteria and only 1 out of the 9 studies demonstrated successful compliance. The most prevalent deviation from the validity criteria was in the fecundity performance (4 out of 9 studies). Although some deviations from the validity criteria were minor, the failure to meet the fecundity criterion cannot be dismissed in a reproduction test where high fecundity is directly related to statistical power. The MEOGRT fecundity validity checks is in principle achievable; however, given the available data, nearly 50% of all studies will need at least one repetition to meet this standard. The high likelihood of study repetition and consequent excessive vertebrate use must be considered when conducting a MEOGRT. The biology of the medaka has advantages, but also poses large hurdles to achieving reliable and valid test results. Therefore, alternative species and/or study designs should be considered to generate the data required to address protection goals, which also reduce the potential for excessive animal use.


Inherent variability in Non-Target Terrestrial Plant (NTTP) guideline testing of pesticides creates challenges in principle achievable; however, given the available data, nearly 50% of all studies will need at least one repetition to meet this standard. The high likelihood of study repetition and consequent excessive vertebrate use must be considered when conducting a MEOGRT. The biology of the medaka has advantages, but also poses large hurdles to achieving reliable and valid test results. Therefore, alternative species and/or study designs should be considered to generate the data required to address protection goals, which also reduce the potential for excessive animal use.

497 An avian reproduction study historical control database: A tool for data interpretation J. Wheeler, Dow AgroSciences; P. Valverde-Garcia, Dow Agro Sciences LLC; T.A. Spencer, EAG Laboratories / Specialist Projects & Histology; V.J. Kramer. Dow AgroSciences LLC / Ecotoxicology; M. Fouldoulakis, Dow Agrosciences / RSRA ERS; I. Barber, Dow AgroSciences

Avian reproduction studies are a regulatory requirement for pesticides in many regions. The data often require careful interpretation due to the nature of the study design, where we present a historical control database. Data from mallard duck reproduction studies performed at the Evans Analytical Group LLC avian toxicology laboratory over the period 1985 - 2016. The analysis demonstrates the stability of reproductive parameters over time and good agreement to normal control ranges as required by the regulatory test guidelines. The major source of variation is shown to be within study variation. Recommendations for the use of historical control data for the interpretation of avian reproduction studies are made. We believe the analysis and evaluation presented here can facilitate the development of practical guidance that can be implemented in regulatory programmes requiring this test.

498 Experimental Design and Model Selection for Ecotox Risk Assessment J.W. Green, DuPont / Data Science and Informatics

Recent experience with regulatory requests for re-analysis of older studies using newer statistical methodology has resurrected an old statistical issue of designing a study to fit its objectives and the dangers of imposing a statistical structure on data not fit for purpose. There is a continual need to update statistical methodology as new ideas arise data is aware to implement these methods become available. Problems can arise when new methods are imposed on old experimental designs. Imagine buying a plot of land with a small cottage. If we tear down the cottage, but leave the cellar and foundation, and then build a mansion in its place but based on the existing foundation, the resulting structure can be unstable and severely restricted in functionality. This presentation will explore the relationship between experimental design and the type of statistical model that can be fit to the resulting data and endpoints that can be estimated or determined from the model. In some instances, newer methods can be applied without problem to existing data. In other cases, existing data cannot support newer methods. It is important to understand the data requirements of the methods or models we intend to use. The size effect that can be estimated or detected is critically important and is strongly related to experimental design and biological variability. There is a model underlying every statistical test used to derive a NOEC or estimate an ECx. The basic statistical model for a simple toxicity experiment is given by Y=µ+ε, where µ is the expected mean response in the 1st concentration, and the ε are independent random errors, usually assumed to be identically distributed. What distinguishes one model from another are what distribution is assumed for the errors or responses and what restrictions or assumptions are placed on the treatment means, µ. It is possible to determine the size effect that can be estimated or detected from a given dataset and it depends largely on experimental design and response variability. Statistical models used for hypothesis testing or regression estimates have data requirements. Model assessment tools are well established and should be used in fitting models to ecological data. Ignoring these tools or model requirements can lead to poorly estimated effects and misleading results. Understanding these concepts enables the scientist to make sound assessments of the data collected.

LCA and beyond - integrating sustainability and/or other dimensions to improve decision support (II)

499 Integration of Risk Assessment and Life Cycle Assessment in the context of recycling wood waste into particleboard S. Huysveld, R.A. Alvarenga, J. Dewulf, Ghent University / Department of Sustainable Organic Chemistry and Technology

Recycling of wood waste into particleboard has some environmental advantages, for instance, by giving second life to wood waste, (i) the use of fresh wood is avoided and (ii) it allows an extended carbon storage, which is beneficial in terms of climate change. However, wood waste sources have a varying chemical quality, therefore, recycling of post-consumer wood waste requires special attention to the determination of chemical composition of the waste. Depending on the type of waste material (e.g., sawdust), for example, may contain high concentrations of heavy metals such as arsenic and copper. Heavy metal toxicity is a threat to the environment and is associated with adverse health effects. In the particleboard industry, heavy metals may be discharged into the air when dust from wood waste is incinerated to supply heat for dryers. Moreover, downstream industrial customers of particleboard (e.g. furniture manufacturers who install the wood particles for internal heat supply), are of concern as well. Local human health effects due to reduced local air quality may question the overall benefit of recycling contaminated wood waste
into particleboard. A need exists to investigate the local human health risks associated with recycling contaminated wood waste, while simultaneously considering other impacts on human health and the environment throughout the entire life cycle traditionally modelled with Life Cycle Assessment (LCA). The objective of this study was the combined use of local Risk Assessment (RA) and LCA to achieve a broader assessment of the sustainability of recycling contaminated wood waste into particleboard. The current scenario, in which the use of contaminated wood waste in particleboard is limited by Flemish government’s standards, is compared to a future scenario with a higher use of more contaminated wood waste. As a consequence, in the future scenario, a lower proportion of the contaminated wood waste will be incinerated with electricity (and heat) recovery. Modeling of the local air pollution is performed with the Immission Prognosis Air Concentration Tool (IMPACT) of the Flemish government. The LCA scope includes the particleboard industry, relevant upstream and downstream processes of the particleboard industry, and the incineration of wood waste. To integrate RA and LCA results on human health effects the concept of disability-adjusted life years (DALYs) is adequate as a common metric. Results will be presented at the conference.

500 Development of non-conventional LCA indicators for circular characteristics of bio-based products

K. Lokked, University of York / Department of Chemistry; X. Bengoa, Quantis; S. Majer, DBFZ, Deutsches Biomasseforschungszentrum gemeinnützige GmbH / Bioenergy Systems; A. Ernstoff, F. Sessa, V. Rossi, Quantis

This paper is dedicated to the identification of key “un-conventional” indicators that demonstrate the sustainability and circular characteristics of promising bio-based products, complementing conventional life cycle analysis. Some of the new LCA complementary indicators proposed as a part of this study emphasised on resource efficiency and material circularity of bio-based value chain and include (but not restricted to) waste circuitry, critical material circularity, land-use efficiency and output quality services. The proposed indicators were drawn out of a comprehensive evaluation of more than 45 certification labels, schemes and initiatives associated to bio-based products, relevant international and national standards and 80+ scientific articles encompassing bio-product life cycle assessment. A current need for a harmonised sustainability certification protocol, coupled with an aim to develop an indicator-led assessment framework lead to the identification of potential gaps in criteria and indicators. Adoption of bio-based products has been identified as the one of the pathways to reach a sustainable economy. Some of the many advantages conceived from adopting bio-based value chains include development of waste-management infrastructure, job creation, SME’s and other environmental opportunities, contributing directly to seven out of 17 UN Sustainable Development Goals. The benefits of such systems approach can be realised only via quantitative and qualitative evaluation of the embedded environmental, techno-economic and societal impacts, all of which are a function of a product’s variables like feedstock type, technology-route, product’s functionality and application [1]. Life cycle assessment, a robust impact-led sustainability analysis that takes into account the stages of a product’s lifecycle using holistic indicators also has insufficiencies, mainly the limitations in addressing the circular product characteristics. This work is a part of the EU-H2020 funded project, Sustainable Transition Assessment and Research of Bio-based products, the ultimate aim of which is to expand existing tools and methodologies for sustainability certification of bio-based products and for their speedy commercial uptake.

501 Toward a more sustainable biochemical industry - Early stage assessments and methodological overlaps between life cycle- and techno-economic assessments of biochemicals

A. Grassland, DTU (Technical University of Denmark) / Division for Quantitative Sustainability Assessment DTU Management Engineering and DTU Biosustain; S. Sukumara, DTU Technical University of Denmark / DTU Biosustain; P. Fantke, Technical University of Denmark / Quantitative Sustainability Assessment Division

Existing Life cycle assessment (LCA) studies of biochemicals reveal that there are challenges that need to be overcome in order to reach an overall high sustainability performance. While in some cases biochemicals have lower global warming impacts compared to fossil-based chemicals, other impacts may become higher, like eutrophication. One of the major sources of environmental impacts of biochemicals is the growing of biomass, which in most cases today is corn. This has led to investment in assessing opportunities of using side streams, like leftover agrovaleral lignocellulose, or in process selection in favour of algae. Macro-algae is one such potential source that given they grow without being farmed, while simultaneously being an important sink for CO2. The objective of this study is to identify trade-offs between assessed environmental impacts and possible burden shifting between macro-algae compared to more conventional feedstocks like maize and lignocellulose. While it is imperative that any change in process configuration reflects in Techno-Economic Assessment (TEA) and LCA, there are very few studies which couples these two assessment demonstrating the trade-offs for improved decision support. The focus of this contribution is to explore methodological overlap between the two assessments and develop a framework, supported by a proof-of-concept. When contrasting current results from the TEA and LCA cradle-to-gate study, some interesting trends were observed. The TEA show that algal biggest hot-spots are identified as feedstock cost which is a function of growing, transportation of biomass and if drying is taking place at the refinery site or closer to the harvesting sites of the feedstock. Whereas, the LCA shows the biggest environmental hot-spots occur in relations to growing of the algae. This requires external application of nutrients and intensive of chemical pretreatment. Today declining costs of chemicals are further developed companies mostly rely of results from TEAs. Our results show that the methodological overlap between TEA and LCA are of that magnitude that justifies the appraisal of this integrated methodology. Introducing LCA as a decision support tool would integrate sustainability requirements in development of technology and solutions. All technologies and products have a life cycle, and by analyzing their impacts, we put numbers on sustainability and benchmark the solutions.

502 A risk evaluation approach for indirect land use change associated to biobased products

D. Marazza, University of Bologna; S. Majer, DBFZ, Deutsches Biomasseforschungszentrum gemeinnützige GmbH / Bioenergy Systems; V. Rossi, Quantis; J. Golazewski, UniversyterWartminskoMazurski W Olsztynie

Biobased products include a vast range of traditional and innovative materials and substances for purposes other than food and energy such as wood and composite materials, bio-plastics, adhesives, lubricants, paints and many other material categories feeding large economic activities. There is international recognition that developing a climate-smart bio-based economy is essential to the continuation of economic development, reduction of greenhouse gas emissions, and adaptation to climatic change. However, as biobased products are ultimately obtained from land or sea, a specific attention has to be payed when considering additional exploitation. Changes of land/sea uses can rebound and cancel out environmental performances and the whole idea of a circular bio-based economy. Indirect impacts of another system have been defined as an unintentional, negative, displacement effect of commodities in the primary sector such as agriculture causing additional land use changes. Provided that ILUC depends on specific legacy effects stemming from land condition prior and after land use changes, overall indirect effects are connected to the 1.1 billion tons of greenhouse gases generated because of land use changes. However, the application of ILUC provisions as for biofuels has been and stays controversial. The Project STAR-ProBio is a multi-actor collaborative research and innovation action and supports the European Commission in the full implementation of European policy initiatives, including the Lead Market Initiative in bio-based products, the industrial policy and the European Bio-economy Strategy. One of the specific goals calls for identifying and mitigating the risks of negative ILUC effects associated to production routes for bio-based products. In this contribution the authors present the conceptual model and the results of the identification of risk factors obtained from the analysis of economic models and a sensitivity analysis performed over one selected case study.

503 How to find sustainable applications for new materials and how to overcome the relativity of LCA

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The Multi-Perspective Material Selection (MPMAS) method has been developed as a decision support tool to find sustainable applications in fields for new materials that are still under development. This selection includes a 3-step method considering technical, economic as well as environmental criteria. So far, the method was best used for new materials that are replacing existing materials in a given application. But applying the MPAS in the case of a completely new kind of material or application field with no clear and existing competitor for comparison, the method reveals its limitations. Especially, the environmental assessment, that uses simplified LCA studies, is a relative approach. Another difficulty for the simplified LCA studies is when the production data of the material and, at the same time, the knowledge about the properties of the end-product are unknown and highly speculative. This is a common problem since the MPAS method is intended to be used after the product or material is technically defined and expands of the MPAS method is made to Step 3, the environmental assessment. The method reveals its limitations meaning that the environmental assessment can be performed without a comparison case and also without the necessity of a lot of data. The development and expansions of the MPAS method is applied to each of its three steps. However, the main development of the method is made to Step 3, the environmental evaluation of the material. Our solution here uses a highly flexible set of criteria that are specifically adapted to the various cases and that are mainly LCA based. This means that the environmental score can now be obtained regardless of the ability to estimate the production data of the material and of the knowledge about the exact properties of the end-product. This evaluation can be applied absolutely or relatively/comparatively. Furthermore, the criteria are expanded with other criteria that go beyond only LCA relevant aspects and also include aspects like circular economy. The method is illustrated with a case study on nanoporous carbonaceous material. As a result, the most sustainable applications for this nanoporous carbonaceous material are identified and used to set parameters to be achieved for
the developers of the material. The new independent environmental assessment part in Step 3 overcomes the necessity of a comparison case while also reducing the required amount of LCA data. This makes the method universally applicable. 

### 504 Consumption and consumer footprint: LCA as pivotal methodology for assessing consumption patterns and ecocivilizations

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The European Commission has been developing an assessment framework to monitor the evolution of environmental impacts associated to the EU consumption. The assessment framework aims at supporting a wide array of policies, such as those related to bioeconomy, resource efficiency, ecocivilization and circular economy. The assessment framework is composed of two sets of consumption-based indicators: the Consumption footprint and the Consumer footprint. The Consumption footprint assesses the potential environmental impact of apparent consumption, focusing on a territorial scale and accounting for trade, assigning the impact to the country where the final consumer is located. The Consumer footprint assesses the potential environmental impact of consumption, based on the results of life cycle assessment (LCA) of representative products purchased and used in one year by an EU citizen. The Consumer footprint allows assessing environmental impacts along the products life cycle (raw material extraction, production, use phase, re-use/recycling and disposal). For the calculation of the Consumer footprint, the consumption of European citizens is split into five key areas (food, housing, mobility, household goods and electric/electronic appliances). For each area, a respective Basket of representative Products (BoP) has been built based on statistics on consumption and stock of product. For each of the five BoPs, a baseline scenario is defined, taking as reference the consumption of an average EU citizen in the baseline year 2010. For the BoP developed for food, the baseline scenario has been modified by selecting foods for identifying the environmental hotspots along the product lifecycle and within the consumption area of each specific BoP. The results of the hotspot analysis are then used as a basis for the selection of actions towards environmental burden reduction, covering either consumption pattern, behavioral changes, implementation of eco-solutions, or a combination of the previous. For each of the actions, a scenario has been developed, by acting on the baseline model and simulating the changes associated to the specific intervention. The LCA results of each scenario are then compared to the results of the baseline, to identify potential benefits or impacts coming from the implementation of the solution tested, as well as to unveil possible trade-offs.

### 505 Environmental Risk Assessment in Sediments

**505 Assessment of risk from historic contaminants in sediments of the Elbe floodplain, using a multiple line of evidence approach**

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The Elbe river has one of the largest catchment areas in Europe (150 000 km²) of which two thirds lie in Germany. While it was once considered to be among the most polluted rivers, water quality has improved since the fall of the Iron Curtain in 1989. Sediments, however, still carry the memory of an industrial past and are often “on the move”, transported by the current and especially during flood events to downstream sites. The question, where they originate, what chemicals they carry and how much of it may still be around, has been in the focus of several previous studies (e.g. Heise et al. 2008, Hillebrand et al. 2015). Little attention, however, had been paid to long-term to sediments in those areas. 1000 backwaters and flood plain lakes along the Elbe with regard to their contamination, their ecotoxicity and their mobility in times of high water discharges and flood plain submergence. Two studies, carried out in 2013 and 2014, were dedicated to this kind of structures with the task of evaluating a potential risk from these sites. Over a stretch of 250 km along the Middle Elbe, sediments from 25 backwaters were sampled, analyzed for heavy metals and for Elbe-typical historic contaminants (HCH, HCB, PCB, PAHs, DDDX). Additional lines of evidence in an assessment of risk comprised the thickness of the sediment layer and sediment mobility during flood events. Dating of sediment cores by 14C analyses facilitated interpretation of the results. Samples were ecotoxicologically tested for inhibitory effects in the bacterial sediment contact test (Arthrobacter globiformis), the luminescence bacteria test (Allivibrio fischeri) with elutriates and methanol extracts and the algae growth inhibition test (Raphidocelis subcapitata) with elutriates. The studies showed that - more than 75 % of all sampled sites were contaminated with heavy metals and organics well beyond the threshold values of the Elbe River Commission. - ecotoxicological effects provided a distinct line of evidence and could not be simply related to analyzed contaminant concentrations. - when integrating chemical, ecotoxicological and erosion stability data into a weight of evidence approach, high risks could be identified for 50 % of the sampled sites in 2013. - dating of sediment cores from 2014 pointed towards a strong impact of the 4 extreme flood events between 2002 and 2013 on the erosion of highly contaminated sediments from backwaters into the Elbe river.

### 506 Multiple lines of evidence for risk assessment of old sea deposits for ilmenite mine tailings in SW Norway

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Analyzing this mine deposits uses up to 3 mill. tons of tailings contaminated with trace amounts of Ni and Cu sulfides. During 1960-94 the tailings were placed in two sea deposits, first in a sheltered fjord and then in a more exposed basin. After 1994 the tailings have been placed in a land-deposit. To protect the downstream watershed area, some of the metal contaminated drainage water is recycled, mixed with other discharge and fed into the water column at the site of the fjord deposit. In 2015 the deposit and reference sites were sampled for studies of macrobenthic communities, biogeochemical fluxes, metal mobilization and metal uptake in gastropods and DGT probes. O₂ and pH in the sediments were measured using micro-electrodes. The tailings were easily traced in the sediments by high concentrations of fine fractions, Fe, Ni, Cu and Co. Tailings were still abundant in the top 0-1 cm of the sediments at both deposit sites, but clearly less abundant at the sediment surface than in deep deposit layers and also less in the outer basin deposit than in the fjord deposit. Compared to Norwegian and European quality standards [1], Cu exceeded MAC-EQS (“Maximum Admissible Concentration”) for coastal sediments indicating a “risk of acute toxic effect” on marine organisms. The DGT-profiles showed that Fe and Mn was recycled within the sediments, whereas Ni and Cu leaked to the overlying water from mobilization centers consistently located in the outer basin deposit. In addition, Cd and organic carbon concentrations were generally lower in the fjord site than in the sea deposits corresponded to 5% of the leakage from the land deposit and 8% of the current discharge to the water-column at the fjord deposit site. Organic carbon (TOC) and fluxes of O₂ and nutrient species were low throughout the investigated area, and the macrobenthic communities showed reduced number of species at the inner fjord site, but ecological status was classified as “good” at all sites and “very good” at the reference site. The statistical test (DGT marginal test) showed that in addition to depth, fine fractions (< 63 µm) and Cu were the only significant environmental parameters explaining the variance in the benthi community data. We conclude that both the current discharge to the water column and the leaching of Cu and Ni from the sea deposits are likely to contribute to the moderate reduction of benthi biodiversity at the old deposit sites. [1] Guideline M-608, 2016. Norwegian Environmental Agency. 24 pp.

### 507 In situ metal fluxes for the assessment of metal bioavailability in sediments

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Analysis of pore waters, dilute-acid extractable metal (AEM), acid volatile sulfide (AVS), and organic carbon (OC) concentrations are frequently used to evaluate and predict metal bioavailability in sediments. Where concentrations of bioavailable contaminants are determined to exceed sediment quality guideline levels, bioassays are usually performed to evaluate toxicity effects resulting from contaminant exposure. Although the chemical analyses used for bioavailability assessment have been shown to be useful for predicting metal toxicity in sediments, 1) the predictions for early oxidized surface sediments can be quite poor, frequently owing to a broader range of factors influencing metal bioavailability including variability in phases that are easily oxidized or reduced (e.g. AVS and Fe(II)). In addition, laboratory-based bioassays may provide inadequate predictions of metal bioavailability and toxicity due to their inability to adequately replicate field exposure conditions. A comprehensive series of studies combining laboratory and field experiments were carried out to evaluate the performance of the diffuse gradients in thin films (DGT) technique for predicting metal bioavailability in sediments. 2) The DGT device uses an ion-exchange resin (Chelex) which selectively accumulates divalent metal present in the sediment porewater and weakly-bound to the sediment particulate phase. 3) The DGT metal flux measured at the sediment water interface (SWI) was compared to biological responses of organisms exposed to sediments contaminated with mixtures of metals, in the laboratory (amphipods and bivalves) and in the field (bivalves). To assist in the analysis of effects from the mixtures of the metals (Cd, Cu, Ni, Pb, Zn), DGT metal fluxes were normalised using water quality guideline values to account for predicted differences in the toxicity of the different metals. Strong dose-response relationships were found between normalised DGT metal fluxes measured at the SWI (DGT) and adverse effects to reproduction and survival of the amphipod exposed to laboratory conditions. Useful predictions of bioaccumulation were obtained for both marine and freshwater bivalves in laboratory and field set-ups. Differences in bioaccumulation between organisms exposed to identical sediments in laboratory and field set-ups highlighted the importance of including in sediment quality assessment lines of evidence based on in situ evaluations of metal bioavailability. 508 An Overview of the Refinements and Improvements to the USEPA’s Sediment Quality Guidelines (2001)
Toxicity Methods for Freshwater Sediment

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Sediment toxicity tests are used for contaminated sediments, chemical registration, and water quality criteria evaluations and can be a core component of ecological risk assessment at contaminated sediment sites. Standard methods for conducting sediment toxicity tests have been established by USEPA, ASTM, Environment Canada and OECD. Revisions to USEPA’s Methods for Measuring the Toxicity and Bioaccumulation of Sediment-Associated Contaminants with Freshwater Invertebrates is planned for 2018. USEPA’s manual describes toxicity and bioaccumulation methods for 3 freshwater species, Hyalella azteca (amphipod), Chironomus dilutus (midge) and Lumbricus variegatus (oligochaete) and 5 sediment toxicity test methods: 10-d tests with H. azteca and C. dilutus; a 42-d life-cycle test with H. azteca; a 50-d life-cycle test with C. dilutus and a 28-d bioaccumulation test with L. variegatus. While laboratories routinely met test acceptability criteria (TAC) for short-term sediment toxicity exposures (10-d control survival and ash-free-dry weight), laboratories reported variable biological performance with the longer exposures. With input from both midge and amphipod workshops, each test method has updated guidance for the starting size/age of organisms, diets and rations, reconstituted water changes, modifications to the test acceptability control survival, weight and other endpoints. Control waters need to have a minimum level of chloride and bromide of a river sand substrate group such as the organisms, Tubifex tubifex, Chironomus riparius, or the amphipod Hyalella azteca, are proposed as Tier-1 test organisms for fungicides with a biocidal mode-of-action. To investigate i) the potential difference between the use of field-collected and artificial sediments and ii) whether the proposed Tier-1 approach is protective, 28-d tests with fluorofoxin-spiked field-collected and artificial sediment were performed with all sediment-dwelling invertebrate taxa individually. Overall, tests for each species and method, laboratories would report their averages for the test endpoints/measures. These proficiency criteria would not be used to accept or reject individual tests, but serve as a broad indicator of laboratory performance and possibly provide insight where refinements are needed. In this talk, we will focus on the testing methods and improvements that have been made in each method for USEPA and ASTM methods. This abstract does not necessarily reflect the view of the policies of the USEPA in

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Sediment-spiked toxicity tests with benthic macro-invertebrates and the fungicide fluorofoxin

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In the EFSA scientific opinion on sediments, one of the oligochaete worms Lumbricillus spp. or Tubifex tubifex, supplemented with a second standard test species Tubifex tubifex, and artificial sediment were performed with all sediment-dwelling invertebrate taxa individually. In a previously performed spiked sediment outdoor microcosm experiment with fluorofoxin the most sensitive sediment-dwelling organism was the oligochaete Dero digita. The test systems used were 1.5L glass vessels containing approximately 2 cm sediment and 1 L aerated spring water. Tests were considered valid if control mortality did not exceed 20% or, in the case of the larva of the midge C. riparius, if more than 70% emergence had occurred. Fluorofoxin concentrations were measured at the start and end of the testing. Endpoints for both worms comprised surviving animals and their weight, including yield and growth rate. For Hyalella both survival, weight and length were assessed, while for Chironomus emergence and total survival was monitored. All tests met the validity criteria of less than 20% control mortality or more than 70% emergence, with the exception of the C. riparius test on artificial sediment. Overall, tests with field-collected sediment gave better results than with artificial sediment. Apparently growth of the animals was better here, allowing for a better expression of effects. Sometimes a factor of 10 difference between the estimated 28d-EC10 value and its upper or lower confidence limit was present, indicating a high associated uncertainty. The confidence intervals were considerably smaller in corresponding 28d-EC0 values, indicating that these are more reliable. In the outdoor microcosm testing the most sensitive organism was an oligochaete. In general, biomass reacted more sensitive than length and survival endpoints. Using either field-collected or artificial sediment data to derive a Tier-1 RAC seems to be sufficiently protective when compared to the outdoor microcosm response.

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Spatial-temporal exposure of Plant Protection Products in OECD 219 sediment test systems - Comparison of model results with measurements

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Sediment toxicity testing among other ecotoxiclogic tests is currently revised under the premise to improve quality and consistency of regulatory environmental risk assessment. In 2015, the European Food Safety Authority (EFSA) has published a scientific opinion on the development of sediment organisms where a water-spiked test system (OECD 219) is considered to study chronic effects on sediment organisms. Prominent test organisms are Chironomids, aquatic insects which live in and on soft sediments. Due to the design of this study initially large gradients between the exposure in the overlying water and in the sediment layer are established. As a consequence, substantial temporal and spatial dynamics of local concentrations have to be expected, especially in the vicinity of the interface between water and sediment where the Chironomids are supposed to stay. To describe local concentrations in such water-sediment test systems we simulated the transport and the redistribution of two moderately mobile (KOC 200 to 300) plant protection products with the mechanistic model TOXSWA. The results of this simulation are compared with measured sediment concentrations in three depths (see contribution submitted by Dorn et al.). The compound properties were parameterised using values derived independently in standard tests (KOC, DT50(water/sediment)) or from literature (diffusion coefficients). Other parameters were derived from OECD 219 experimental design information. The simulations matched the measured concentrations spatially and temporally well. The simulated concentration depth profiles averaged for the layers which were measured lay almost always within the range of single measurements. Also the concentrations in the overlying water were reproduced well. The main findings are that the concentrations in the sediment show a pronounced temporal pattern and that the concentrations in the sediment are strongly depth-dependent. The dominant transport process in the sediment is obviously diffusion which however did not lead to homogeneous penetration of the sediment. Presuming that chironomids live in the upper three millimetres of the sediment, they are exposed to approximately four times higher test compound concentration (total and liquid) than the average concentration in the sediment. This has important consequences for the derivation of effect endpoints of chironomids from OECD 219 study data, which should consider the local exposure of the organisms.

Wastewater effluents: How research can improve risk assessment and regulation

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Effects of untreated wastewater dilution in surface waters on pharmaceuticals natural attenuation and on the community genomics: Implications for ERA

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The decreasing consumption and production of active pharmaceutical ingredients (APIs) in low and low-middle income countries (LMMICs) is of growing environmental concern owing to their possible ecotoxicological effects. This is related to the practice of direct discharge of untreated wastewater (DWW), which creates a heavily polluted area, named the “impact zone”. Little is known about the environmental fate of APIs in this area. Nevertheless, a few available measured environmental concentrations (MECs) of LLMICs show higher concentrations than for high-income countries with developed wastewater treatment infrastructures. Globally, the MECs of APIs in the “impact zone” are typically above 0.01 µg L⁻¹, which, if predicted, would trigger the environmental fate of APIs in the environmental risk assessment (ERA). In the ERA PEC calculation, a default dilution factor (DF) of 10 is used, but in at least 53 countries worldwide, the local environmental concentrations (MECs) of APIs mixture and the dilution on the microbial communities is missing, hence necessitating the requirement for evaluation of endpoints for high-consumption areas. Such gaps are present in the literature about the effects of low dilutions on the natural attenuation of APIs or impacts of DWW. Furthermore, information on the effects of low dilution on mixtures of APIs is missing, hence necessitating the requirement for evaluation of ecological endpoints for the impact zone ERA. This information is pivotal for the development of an impact zone ERA approach, and we are proposing an original approach to expand this area of concern, which shows persistancy but the concentration is below the sensitivity of the methods used. Accordingly, in this contribution we present possible pathways for the fate of APIs in wastewater effluents: How research can improve risk assessment and regulation.
Wastewater Treatment Works: Measurement, Prediction, Risk - A Cause for Concern?

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This work reports on the ability for wastewater treatment works (WwTW) to remove active pharmaceutical ingredients (APIs), the variations within and between works, the effectiveness of trying to model removal and the risk of exceeding predicted no effect concentrations (PNEC) in the environment. The research is based on data generated from two large UK-wide WwTW monitoring programmes. Taking account of removal of parent compound from the aqueous phase during treatment in combination with estimates of dilution available it is possible to prioritise the APIs of greatest risk of exceeding estimates of their PNEC in receiving waters for all WwTW in the UK. The majority of substances studied were removed to a high degree, although with significant variation, both within and between WwTW. Poorer removal (between influent and effluent) was observed for ethinylestradiol, diclofenac, propranolol, the macrolide antibiotics, fluoxetine, tamofoxen and carbamazepine. All but the last two of these substances were present in effluents at concentrations higher than their respective estimated PNEC (based on measured parent concentrations from 45 WwTW on 20 occasions). The application of models to predict removal efficiencies are reported. Based on available dilution data as many as 890 WwTW in the UK (approximately 13% of all WwTW) may cause exceedances of estimated riverine PNECs after mixing of their effluents with receiving waters. The overall degree of risk is driven by the toxicity value selected, which in itself is controlled by the availability of reliable and relevant data. Part of the project focuses on assessing the health status of gammarids and the macrozoobenthos community in the Schussen river. Samples were taken up- and downstream of the WwTW, as well as before and after the upgrading of the WwTW. Gammarid populations from all sites were investigated with respect to sex ratio and fecundity of breeding females. In addition, analyses of heat shock protein (hsp70) levels and lipidperoxides allowed us to draw conclusions about protozoic and oxidative stress in gammarids. Macrozoobenthos community integrity was determined by means of the saprobic index as well as by the number of sensitive taxa (EPT index). Prior to the WWTP upgrade, the health status of gammarids as well as the integrity of the macrozoobentho community was determined by molecular biology techniques. The results highlight a promising tool for protozoa depuration. Keys words: protozoa, wastewater treatment plant, bivalve, depuration

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SETAC Europe 28th Annual Meeting Abstract Book

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Pharmaceuticals have been found in aquatic systems globally, due to a combination of worldwide usage and low removal efficiency in wastewater treatment plants (WWTPs), or a complete lack of WWTPs (1). In surface waters, concentrations of pharmaceuticals usually range from low ng l⁻¹ to close to point sources to low ng l⁻¹, and are correlated to human population density in the drainage area, volume of the receiving water body and presence of WWTPs. One technique to remove the removal of pharmaceuticals in WWTPs is to add a tertiary treatment step based on the addition of ozone. Ozone is a cost efficient way to degrade chemicals and several studies have shown that most pharmaceuticals are readily degraded in the presence of ozone (2). However, several oxidized degradation products are formed during ozonation and the environmental impact of these are largely unknown. The aim of this study was to investigate the environmental impact of ozonation in a WWTP, when adding ozonation as an additional tertiary treatment step and to investigate the environmental impact of this effluent on the receiving river. All treated effluent from a minor WWTP (10000 PE) were treated by an addition of 8 h⁻¹ ozone during 6 months. Removal rates in the WWTP as well as levels of pharmaceuticals in the receiving river (both in water and biota) were monitored. Surface water data from 10 sampling sites and 10 sampling occasions before, during and after ozonation, will be presented. Ecological status and levels of pharmaceuticals in exposed biota (n=5) at each site and sampling occasion also will be presented. Several additional methods to evaluate the impact of ozonation was used including impact on microbial community composition, presence of antibiotic resistance genes as well as studies to detect endocrine, reproductive and behavioral effects in fish and its progeny.

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Effects of full scale ozonation of treated effluent - Environmental impact in a receiving river

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Dreissena polymorpha as a purification tool for protozoa in wastewater treatment plant effluent

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This work reports on the ability for wastewater treatment works (WwTW) to remove active pharmaceutical ingredients (APIs), the variations within and between works, the effectiveness of trying to model removal and the risk of exceeding predicted no effect concentrations (PNEC) in the environment. The research is based on data generated from two large UK-wide WwTW monitoring programmes. Taking account of removal of parent compound from the aqueous phase during treatment in combination with estimates of dilution available it is possible to prioritise the APIs of greatest risk of exceeding estimates of their PNEC in receiving waters for all WwTW in the UK. The majority of substances studied were removed to a high degree, although with significant variation, both within and between WwTW. Poorer removal (between influent and effluent) was observed for ethinylestradiol, diclofenac, propranolol, the macrolide antibiotics, fluoxetine, tamofoxen and carbamazepine. All but the last two of these substances were present in effluents at concentrations higher than their respective estimated PNEC (based on measured parent concentrations from 45 WwTW on 20 occasions). The application of models to predict removal efficiencies are reported. Based on available dilution data as many as 890 WwTW in the UK (approximately 13% of all WwTW) may cause exceedances of estimated riverine PNECs after mixing of their effluents with receiving waters. The overall degree of risk is driven by the toxicity value selected, which in itself is controlled by the availability of reliable and relevant data. Part of the project focuses on assessing the health status of gammarids and the macrozoobenthos community in the Schussen river. Samples were taken up- and downstream of the WwTW, as well as before and after the upgrading of the WwTP. Gammarid populations from all sites were investigated with respect to sex ratio and fecundity of breeding females. In addition, analyses of heat shock protein (hsp70) levels and lipidperoxides allowed us to draw conclusions about protozoic and oxidative stress in gammarids. Macrozoobenthos community integrity was determined by means of the saprobic index as well as by the number of sensitive taxa (EPT index). Prior to the WWTP upgrade, the health status of gammarids as well as the integrity of the macrozoobentho community was negatively influenced by the WWTPs effluent. After the upgrading of the WwTP, gammarids from the downstream site did not differ any longer from those collected upstream of the WwTP with respect to the investigated health parameters. Furthermore, the overall number of taxa and particularly the number of EPT taxa within the macrozoobenthos community downstream of the WWTP increased distinctly after the upgrade of the WwTP with the additional activated carbon step. We conclude that the efficiency of the activated carbon step to eliminate toxic and endocrine active substances from the effluent could plausibly be related to the improved integrity of macroinvertebrate health and community structure in the connected river Schussen.
used to water supply and has been reported as contaminated by cyanotoxins and pharmaceuticals. Therefore, this contamination increases costs to the water treatment and can cause toxic effects to the aquatic organisms and human health. The aim of this study was to test Green Liver System to remove the contaminants, at the same concentrations that were found in the reservoir, using aquatic macrophytes. *Egeria densa*, Ceratophyllum demersum and Myriophyllum aquaticum were exposed to concentrations of paracetamol, diclofenac and microcystin-LR using a laboratory model of the Green Liver System for 14 days. Water samples were collected in 0, 1, 3, 7 and 14 days and plants samples were collected at the end of the experiment. Two control experiments were carried out in parallel. Water and plants samples were used to quantify the contaminants and plants samples were also used to evaluate the catalase and glutathione S-transferase activities. Plant species took up the contaminants and the removals of compounds were 93% for diclofenac and 100% for microcystin-LR. Our results showed that the plants antioxidant system was not activated and the Green Liver System was a suitable methodology to clean the water and to implement in phytoremediation programs. **Keywords:** Green Liver System, Reservoir, aquatic macrophytes, phytoremediation.

**Antibiotics and Antibiotic Resistance in the Environment: Fate and Ecological Effects, Resistance Development and Implications for Human Health**

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**Identifying hotspots of Antimicrobial Resistance Selection in the Natural Environment**  
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Releases of antimicrobials into the environment increase selective pressures on environmental microbes contributing to the proliferation of antimicrobial resistance (AMR) and perhaps the inevitable ‘post antibiotic era’. One of many challenges in understanding the environmental and organismic factors required to provide widespread environmental monitoring. In lieu of this monitoring data, predicted environmental concentration (PEC) modelling based on pharmaceutical usage data has been demonstrated to be a useful tool in approximating antimicrobial exposures to the environment. Recently, attempts have been made to predict no-effect concentrations (PNEC) for selective pressures in the development of AMR. Coupling PEC and PNEC values provides a powerful tool to estimate the risks associated with a particular compound or class of compounds relating to AMR proliferation. Here we use this approach to identify hotspots where antibiotic exposure may be contributing to AMR selection for a range of different scenarios. Antibiotic usage data, data on metabolism, wastewater treatment and dilution data were used to determine PEC values, which were compared with reported PNECs to determine AMR hotspots for 56 compounds used in Wales as well as 9 chemical classes of antimicrobials in European Countries. Finally, using daily flow data, the approach was applied to a single wastewater treatment utility serving a population of approximately 18,600 persons with effluent discharge into the River Foss, UK to highlight the variation patterns in daily risk associated with AMR selection. Having illustrated the utility and approach for a range of spatial and temporal scenarios, we believe that these results will be invaluable in informing future monitoring of antibiotics and AMR in the environment.

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**Urban and rural antibiotic resistance**  
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Soils are both a source and a sink for antimicrobial resistance (AMR). Despite growing awareness of AMR in the soil resistome, debate continues over responsibility for increased AMR dissemination in this important environmental reservoir. While soil AMR is innate, the relative abundance of antibiotic resistance genes (ARGs) in soil has significantly increased over the last 60 years since the release of antimicrobials into the environment. The scale and multi- aspect of antibiotic resistance genes (ARGs) in soil has significantly increased over the last 60 years since the release of antimicrobials into the environment. Antibiotic usage data, data on metabolism, wastewater treatment and dilution data were used to determine PEC values, which were compared with reported PNECs to determine AMR hotspots for 56 compounds used in Wales as well as 9 chemical classes of antimicrobials in European Countries. Finally, using daily flow data, the approach was applied to a single wastewater treatment utility serving a population of approximately 18,600 persons with effluent discharge into the River Foss, UK to highlight the variation patterns in daily risk associated with AMR selection. Having illustrated the utility and approach for a range of spatial and temporal scenarios, we believe that these results will be invaluable in informing future monitoring of antibiotics and AMR in the environment.

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**Dissemination of extending-spectra β-lactamase E. coli carrying multidrug resistance and virulence factors in tropical rivers receiving hospital effluents**  
A. Lufftine, University of Geneva / Institut Forel; V. Slavenskova, University of Geneva / Département F.-A. Forel des sciences de l'environnement et de leau; J. Poté, University of Geneva / Department F.A. Forel of environmental and aquatic sciences

The occurrence and dissemination of antibiotic resistant bacteria and their resistance genes from clinical settings to environmental compartments have become a major concern because of serious threat human health worldwide. Given the serious clinical threat of Extended-spectrum β-lactamases (ESBL) and Carbapenem-resistant Enterobacteriaceae (CRE), studies are available in many countries from clinical settings. However, there is the dearth of studies in environmental compartments for the presence of these high threat gram-negative bacteria. This situation is particularly alarming in developing countries in which the freshwater resources receive urban and hospital effluent water without previous treatment. Additionally, most tropical urban rivers are associated to resistance against numerous antibiotic classes (i.e. inhibitors combination, 4th generation of β-lactams, monobactams, carbapenems, aminoglycosides, tetracyclin, quinolones and phenicol classes) and may also carry virulence genes factors. The prolonged usage of multi-drug resistant E. coli are not being linked to untreated hospital wastewater discharge in urban receiving system and are widely distributed along the river, thus highlighting the risk of surface water use.
watchlist in 2015 due to their measured environmental concentrations (MECs) and predicted environmental concentrations (PECs) being higher than their predicted no effect concentrations (PNECs). The aims of this study were to investigate the selective potential of these three compounds in a complex microbial community and to determine minimal selective concentrations (MSCs) for each. A number of week-long evolution experiments were conducted at a range of macrolide concentrations. QPCR determined the presence of a variety of macrolide resistant genes (ermB, ermC, mbr, msrA, msrD and mefE family) in strain S11 within the community. Change in prevalence of resistance genes, when in the presence of antibiotic, was compared to change in prevalence when no antibiotic was present. Out of all of the genes tested, the ermF gene shows a selective response at the lowest concentration for all three macrolide antibiotics. No significant selection is seen for ermA at 50μg/L, but we do see significant selection at 75μg/L for all three compounds. The highest current MEC for any of these macrolide compounds is 4μg/L (erythromycin-H2O). Our data suggests, therefore, that current environmental concentrations of the macrolide compounds do not select for resistance genes in a complex microbial community.

### 522 Impact of multi-year exposure of agricultural soils to antibiotics on the soil resistome and mobiles.

C.H. Lau, Y. Tien, Agriculture and Agri-Food Canada; E. Topp, Agriculture and Agri-Food Canada (AAFC)

Antibiotics are entrained into agricultural soil through the application of animal manures and sewage sludge. In order to understand the potential long term effects of antibiotics on soil microorganisms, field plots were established in 1999 that have since received annual applications of a mixture of tylosin, chlortetracycline and sulfamethazine, and a second series of plots was established in 2010 that receive a mixture of erythromycin, clarithromycin and azithromycin. Antibiotics have been applied every spring at concentrations ranging from 0.1 to 10 mg kg soil\(^{-1}\), and plots have been continuously cropped to soybeans. A library of large cloned fragments was constructed using DNA sampled in 2014 from plots receiving the highest application rates, or no antibiotics. The library was cloned into antibiotic-sensitive Escherichia coli, and antibiotic resistance genes (ARGs) in the library were discovered by identifying E. coli clones that grew upon plating on solid growth media containing various antibiotics. Genes encoding resistance to many classes of antibiotics including the sulfonamides, tetracyclines, macrolides and \(\beta\)-lactams were identified, and sequence analysis revealed some to be entirely novel (Lau et al. 2017 Appl. Environ. Microbiol. 83 no. 16 e00989-17). A key question was whether the abundance of these genes increased in response to antibiotic exposure, evidence that would be consistent with functional importance in situ. The abundance of the novel targets as well as previously known ARGs, integrons and plasmids in soil DNA was quantified by real time PCR or with the WaferGen Biosystems high throughput qPCR instrument. Some gene targets (e.g. intI1, sul1, strA) were much more abundant in soils exposed to antibiotics whereas the vast majority of targets were not detectably increased in abundance. Overall, these results suggest that genes associated with integron cassettes are amplified in soil following repeated exposure to antibiotics.

### Distribution, transformations and biological effects of incidental nanoparticles and nanoplastics in the environment from a more realistic point of view

#### 523 Inter-annual monitoring of microplastics in marine intertidal sediments of the Firth of Forth

M. Hartl, Heriot-Watt University / Centre for Marine Biodiversity & Biotechnology, Institute of Life and Earth Sciences; Z. Lawrence, Heriot-Watt University / Institute of Life and Earth Sciences, Centre for Marine Biodiversity & Biotechnology; C. Holmes, A. Deery, Heriot Watt University / Centre for Marine Biodiversity and Biotechnology Institute of Life and Earth Sciences; J. Blumenröder, Heriot Watt University / Institute of Life and Earth Sciences, Centre for Marine Biodiversity & Biotechnology; P. Sechet, Heriot Watt University / Centre for Marine Biodiversity Biotechnology Institute of Life and Earth Sciences; R. Wood, Heriot Watt University / Institute of Life and Earth Sciences, Centre for Marine Biodiversity & Biotechnology; M. Mears, Heriot Watt University / Centre for Marine Biodiversity and Biotechnology School of Life Sciences; S. Paterson, Heriot Watt University / Centre for Marine Biodiversity Biotechnology Institute of Life and Earth Sciences; M. Viguiaud, H. Walker, F. Kinsley-Willis, J. McCretton, Heriot Watt University / Institute of Life and Earth Sciences, Centre for Marine Biodiversity & Biotechnology

Microplastics (MPs) and microfibres of plastic smaller < 5mm are commonly found in the marine environment and originate either consumer care products and plastic production plants or from the disintegration of larger pieces. MPs need to be monitored in order to evaluate the effectiveness of Government initiatives to reduce plastic debris in the environment. The aim of the present study, therefore, was to contribute to the development of a hitherto lacking quantitative long-term marine MPs database. We present the results of a three-year pilot project in the Firth of Forth, point to innovations in sampling and contamination prevention, as well as the limitations. Sediment samples were obtained in triplicate from intertidal sites in May2014, May & Sept2015, May & Sept2016, 2017 using glass bijoux tubes as miniature cores and sealed with metal screw caps, processed using a density separation procedure and the polymer types determined using FT-IR spectroscopy. The results showed that there are high numbers of plastic particles (34-8400 kg\(^{-1}\)) and fibres (1,700-4,300 kg\(^{-1}\)) along both shores of the Firth of Forth. The number of Fibres was generally higher than MPs. There was no apparent pattern of spatial distribution. Although a spike in MP particle numbers occurred in September 2015 and May2016, there was no significant difference in MP particle concentrations between May 2014 and May 2017. There was also no significant difference in MP fibre concentrations during the same three-year period. There was also no evidence of seasonal fluctuations in MP concentrations. The results show that, for intertidal sediments in the Firth of Forth, the MP concentration has remained stable. This is significant baseline information and will be instrumental in assessing the effectiveness of Government policy regulating industry and consumer behaviour towards the production and use of particularly single-use plastic products. However, in order to compare results between countries and laboratories, for the purpose of gaining a more global insight into the micropollutants contamination issue, more standardized sampling and extraction procedures need to be developed.

### 524 Do nanoparticles cause stress effects on microalgae? An infrared spectroscopy study.

M. Dénier, Institute of molecules and materials of Le Mans / Physique des Interfaces et des MésoStructures; N. Enrizen, Institute of molecules and materials of Le Mans; A. Caruso, laboratory Mer Molecule Santé; f. lagarde, Institute of molecules and materials of Le Mans / Institute of Materials and Molecules of Le Mans IMM IMUM CNRS

Nanoparticles are constantly used at world level leading to their presence in the aquatic environment lead to possible particles interaction with living organisms. The potential impacts of suspended nanoparticles on aquatic organisms is thus understood of the induced mechanisms. Microalgae are the base of aquatic trophic chain and different possible pathways of interaction between microalgae and nanoparticles are described in literature [1]. Moreover, infrared spectroscopy is known to monitoring material chemical composition. In this work, infrared spectroscopy is used to monitor the interaction between different types of nanoparticles and a model freshwater microalgae (Chlamydomonas reinhardtii). The aim is to understand the effect induced by nanoparticles and discriminate the responses of the microalgae in comparison with known stress mechanisms. Different stresses are realised in this goal: nutrient deficiency, light deficiency, metallic stress, gold and polystyrene nanoparticles. After 48h or 72h of interaction, microalgae are collected for infrared analysis. In parallel, biological parameters towards a representative organism of aquatic ecosystems

### 525 Ecotoxicological evaluation of high-generation cationic PAMAM dendrimers towards a representative organism of aquatic ecosystems

G. Pulido-Reyes, M. Tamayo-Belda, M.G. Pleiter, Universidad Autónoma de Madrid; k.m. betancor, F. Leganes, Universidad Autónoma de Madrid / Biology; R. Rosal, University of Alcala; F. Fernandez-Pitas, Universidad Autónoma de Madrid / Biology

Nowadays, nanomaterials are extensively used worldwide in many different fields and their potentially serious effects in aquatic ecosystems have become a global concern [1]. Poly(amidoamine) (PAMAM) dendrimers are polymeric nanomaterials, radially symmetric, dendritic molecules and materials of Le Mans / Institute of Materials and Molecules of Le Mans IMM IMUM CNRS

Nanoparticles are constantly used at world level leading to their presence in the aquatic environment leading to possible particles interaction with living organisms. The potential impacts of suspended nanoparticles on aquatic organisms is thus understood of the induced mechanisms. Microalgae are the base of aquatic trophic chain and different possible pathways of interaction between microalgae and nanoparticles are described in literature [1]. Moreover, infrared spectroscopy is known to monitoring material chemical composition. In this work, infrared spectroscopy is used to monitor the interaction between different types of nanoparticles and a model freshwater microalgae (Chlamydomonas reinhardtii). The aim is to understand the effect induced by nanoparticles and discriminate the responses of the microalgae in comparison with known stress mechanisms. Different stresses are realised in this goal: nutrient deficiency, light deficiency, metallic stress, gold and polystyrene nanoparticles. After 48h or 72h of interaction, microalgae are collected for infrared analysis. In parallel, biological parameters towards a representative organism of aquatic ecosystems

### 526 Howev...

526 Interactive effects of carbon nanoparticles and benzo(c)pyrene on marine mussels, Mytilus galloprovincialis

A. Barranger, University of Plymouth / School of Biological Sciences; Y. Aminot, University of Plymouth; M. Banni, Laboratory of Biochemical and Environmental Toxicology; S. Sforzini, Universita Del Piemonte Orientale Amadeo Avogadro / Department of Sciences and Technological Innovation (DiSt); V. M. Atil, Kings College London; A. G. Jiang, A. Khlebtsov, University of Nottingham / School of Chemistry; A. Viarengo, University of Piemonte Orientale / Department of Sciences and Technological Innovation DiSt; J.W. Readman, University of Plymouth / Biogeochemistry Research Centre; A. N. Jha, Plymouth University / Biological Sciences.

The production and discharge of nanoparticles has grown extensively over the last few years, raising concerns over their potential impact on environmental health, either alone or in combination with other anthropogenic contaminants. The study, funded by Natural Environment Research Council (NERC), UK aims to test the hypothesis that environmentally relevant carbon based nanoparticles (CNPs) and polycyclic aromatic hydrocarbons (PAHs) can interact with each other to differentially modify their potential toxicity. To probe this hypothesis, marine mussels were exposed for 3 days to benzo(c)pyrene of different types of carbon nanoparticles, [C60 fullerene and multi-walled carbon nanotubes (MWNNTs)], both alone and in combination with BaP. Tissue specific distributions and concentrations of CNPs and BaP were determined in exposed mussels. To enhance the analytical traceability of these CNPs in biological systems, some nanoparticles were labelled with rare elements. CNP uptake was followed by ICP-MS and/or HPLC-UV, with the BaP uptake tracked by GCMS. CNP uptake was also investigated by electron microscopy. The genotoxic effects were characterised by the level of DNA strand breaks (comet assay), micronuclei and DNA adduct analyses. Global gene expression profiles were analysed using microarray technology targeting 15 stress response pathways. Contrasting results were obtained as G. bangsi exposed to carbon nanoparticles used carbon mussels to MWNNTs and BaP seems to reduce the uptake and genotoxic effects of BaP in the digestive gland. Conversely, co-exposure to C60 and BaP does not seem to affect the uptake and genotoxic effects of BaP. Different responses were also observed with the transcriptomic studies. Microarray analysis identified several key biological processes (e.g. DNA metabolism, cytoskeleton, oxidative stress and heat shock factors). In order to have a better understanding of the effects of these CNPs, further biological analysis (e.g. DNA oxidation and proteomics) are currently in progress. This study opens new questions highlighting the importance of studying the potential interaction between nanomaterials and environmentally important pollutants.

527 Trophic transfer of CuO NPs and aqueous Cu: from worms to fish - a proof of concept study

T. Lammel, University of Gothenburg / Dep of Biological and Environmental Sciences; A. Thit Jensen, Roskilde University / ENSPAC; C. Mouneyrac, Université de Lorraine / Dept Science and Environment.

First evidences of PAMAM dendrimer internalization in microorganisms of wastewater bioluminescence. A linkage with the living and oxidative stress. Nanotoxicology 9(6): 706-718 Acknowledgement - This research was supported by CTM2013-45775-C2-2-R and CGL2010-15675 grants from MINECO/FEDER EU.

528 Corbicula fluminea exposure to copper oxide nanoparticles: an integrated mesocosm study

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Engineered nanoparticles (ENP) are now part of our daily life because of their introduction in a wide variety of products. Their concentrations in environment are not yet known but release during their life cycle is obvious. Copper oxide nanoparticles (CuO ENP) are well known for their antimicrobial properties allowing their use in numerous products as in wood-paints, textiles or food packaging. Since aquatic compartments are the ultimate sink of contamination, they should be impacted by release of ENP. Some studies highlighted the ability of CuO ENP to induce stress responses in several levels of biological organisation in aquatic organisms, indicating their toxic potential. Most studies were nonetheless made using simplified exposures, thus maximizing ENP dispersion and contact with the studied organisms. The fate and consequently the toxic potential of CuO ENP differ depending on the complexity of the exposure media that can considerably modify ENP physico-chemical properties and consequently, their bioavailability to living organisms. Thereby, setting up more complex design of exposure may help to gain in environmental realism. The aim of this work was to evaluate the fate and effects of different CuO ENP on a widespread endobenthic freshwater bivalve Corbicula fluminea. In order to improve environmental realism, C. fluminea were exposed in indoor mesocosm containing sediment, water and food. A cumulative contamination was applied until reaching a final concentration of 50 µg CuO/L at the end of the exposure period (28 d). Behavioral, physiological, biochemical and molecular parameters were quantified in order to assess CuO ENP impacts on C. fluminea. Results of this study allow to conclude that CuO ENP affected C. fluminea behavior, ENP bioavailability and accumulation in fish is influenced by the mesocosm treatment. These results suggest that transfer of CuO NP along the food chain may be limited compared to dissolved Cu. We suggest future studies on how NP bioavailability and accumulation in fish is influenced by other important factors, such as exposure dose, time and NP properties upon biotransformation by the foraging organism.

529 Luminescent biomonitoring via bioassays of different complexity - from cells trough enzyme reactions to proteins

V. Kratasyuk, University of Nottingham / School of Biological Sciences.

A new approach in developing bacterial bioluminescent enzymatic biosensors, application to toxicity bioassays, and the needed reagents has been developed. To solve the problem of how to quickly, easily, and accurately measure the numerous chemical compounds in environmental monitoring, food product contamination, and medical diagnostics, bioluminescent enzymatic toxicity assays were prepared, wherein the bacterial coupled enzyme system NAD(P)H:FMN-oxydoreductase-luciferase substitutes for older methods using living organisms. The immobilized reagent Enzymolux was used to facilitate and accelerate the development of the bioluminescent enzymatic biosensors used for biomolecular and biological toxicological assays. The reagent is easy to use and convenient to be applied not only in toxicity studies but also in education, mainly in ecological and enzymological practical courses. Prototype biosensors offer cost advantages, versatility, high sensitivity, rapid response, extended shelf-life and flexible storage conditions. This study was supported by the Russian Science Foundation (project no. 16-14-10115).

530 Toxic and adaptive effects via luminescent assay systems of different complexity - from cells trough enzyme reactions to proteins

V. Kratasyuk, University of Nottingham / School of Biological Sciences.

A new approach in developing bacterial bioluminescent enzymatic biosensors, application to toxicity bioassays, and the needed reagents has been developed. To solve the problem of how to quickly, easily, and accurately measure the numerous chemical compounds in environmental monitoring, food product contamination, and medical diagnostics, bioluminescent enzymatic toxicity assays were prepared, wherein the bacterial coupled enzyme system NAD(P)H:FMN-oxydoreductase-luciferase substitutes for older methods using living organisms. The immobilized reagent Enzymolux was used to facilitate and accelerate the development of the bioluminescent enzymatic biosensors used for biomolecular and biological toxicological assays. The reagent is easy to use and convenient to be applied not only in toxicity studies but also in education, mainly in ecological and enzymological practical courses. Prototype biosensors offer cost advantages, versatility, high sensitivity, rapid response, extended shelf-life and flexible storage conditions. This study was supported by the Russian Science Foundation (project no. 16-14-10115).
The possibilities of using fungal fluorophores to assess responses of filamentous fungi to external stimuli

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Ecotoxicological Soil Analysis; M. Freidkin, Lomonosov Moscow State University / Department of General Physics; S. Patsaeva, Lomonosov Moscow State University / Department of General Physics

The interest to functional and structural indicators of mycobacteria with a respective to use them in biogagnostics is currently rising because fungi represent as essential component among environmental decomposers of organic material in ecosystems. The complexity of utilization of fungi in biogagnostics is explained by the variety of reactions to external stimuli, in their physiological and biochemical plasticity. In this regard, it seems very important to find the reactions of fungal cultures adequately reflecting their response to external stimuli in different conditions. The essential biogenic fluorophores such as NADH, tryptophan, melanin, ergosterol, pyridoxine, riboflavin, FAD, and FMN can be monitored instrumentally by spectroscopic techniques. The aim of the present study was to investigate the features of fluorescence spectra of fulamanous fungi cultivated under different concentrations of source of bioavailable and nor readily bioavailable carbon in the growth medium. The research objects were strains of Alternaria alternata, Cladosporium cladosporioides, and Trichoderma harzianum. The strains were kindly provided by O.E. Marfenina and A.E. Ivaniva, Soil Science Faculty of MSU. The filamentous fungi Staphylococcus aureus and agar Czapek medium contained a varying level of sucrose (0.3% and 3%) and humic substances (0.02 and 0.1%). Fluorescence spectra were measured using a luminescence spectrometer Solar CM2203 at several wavelengths of the exciting radiation (250, 280, 310, 350, and 450 nm). Fluorescence excitation spectra were registered for emission at 350, 440, and 460 nm. Typical fluorescence spectra of fungal samples (sporangia of powd and NP's original- and engineered UV-excitation consist of two overlapping bands. The UV-band with maximum at 300-350 nm under excitation at 280 nm is a protein-like fluorescence, and the band in the blue region with the maximum at 400-450 nm under excitation at 310-370 nm is emission of fungal chomophores like NADH and or melanins. We suggest using the intensity of the first fluorescence band in the fungal samples for rapid evaluation of spare biomass. Measured fluorescence characteristics were found correlating with saturation of growth medium by source of bioavailable and not readily bioavailable carbon. Therefore we consider this research as promising on the way of using fungal fluorophores to assess responses of filamentous fungi to external stimuli.

Poster spotlight: WE209, WE210, WE211

Obesogens and lipid disruptors

SETAC Europe 28th Annual Meeting Abstract Book
534 The evolution of obesogen-induced phenotypes in vertebrates
A. Cañada, M. Lopes-Marques, R. Ruivo, E. Fonseca, R. Jorge, M. Barbosa, CIMAR - University of Porto; Y. Hiromori, Y. Ishi, T. Miyagi, T. Nakamish, Gifu Pharmaceutical University; R. Santos, Hepia University of Applied Sciences Western Switzerland; E. Castro, CIMAR - University of Porto

Global obesity is an escalating pandemic in western societies. Triggered by numerous environmental and behavioral factors, this epidemic condition is also influenced by individual and environmental cues. Of note are the globally persistent man-made chemicals, with ever-growing ecosystemic consequences, a hallmark of the Anthropocene epoch. A striking example highlights the role of a group of compounds known as “obesogens”. In mammals, most examples involve the modulation of the peroxisome proliferator-activated receptor γ (PPARγ) nuclear receptor. To decipher the pattern of PPARγ exploitation by a model obesogen, tributyltin (TBT), we employed an extensive analysis from comparative genomics to transactivation assays, site-directed-mutagenesis, and homology modeling, to unfold the structural and biological determinants of PPAR exploitation by TBT. Our findings endorse the modulatory ability of man-made chemicals and suggest an evolutionary diverse setting of “obesogenic” responses to TBT, with impacts for human health risk assessment.

536 Aging Extension and Modifications of Lipid Metabolism in the Monogonont Rotifer Brachionus koreanus under Chronic Caloric Restriction
M. Lee, J. Paek, J. Lee, Sungkyunkwan University

To examine the relationship of the aging extension and modification of lipid metabolism under chronic caloric restriction (CCR; concentration from 0 to 100% of the diatom Tetraselmis suecica) in the monogonont rotifer Brachionus koreanus, we assessed the life cycle parameters, fatty acid composition, and sirtuin and lipid metabolism-related genes. As a result, in the 5% exposed group, B. koreanus showed the decreased life span and less fecundity compared to the control. Based on this finding, we chose 5% of T. suecica and performed the rest of the experiment compared to 100%. As a result, up-regulation of sirtuin genes expression was observed. In addition, despite the reduction in the amount of total fatty acid (FA) and the area of triacylglycerol, the increase in the ratio of saturated fatty acid and monounsaturated fatty acid (MUFA) among the total FA in 5%–exposed B. koreanus were observed. Furthermore, the mRNA expression of Δ9 desaturase confirmed that CCR promoted the synthesis of MUFA through Δ9 desaturase. Moreover, the expression of docosahexaenoic acid (DHA) synthesizing gene, Δ6 desaturase, has also been up-regulated along with DHA content. These data suggest that CCR modified histone acetylation and lipid metabolism, leading to decrease in reproduction, consequently resulting in life span extension.

537 Lipidomic and transcriptomic changes induced by compounds enhancing accumulation of storage lipids in Daphnia magna.
I. Fuigues, Institute of Environmental Assessment and Water Research IDAEA CSIC; R. Juidao, Institute of Environmental Assessment and Water Research IDAEA CSIC / Department of Environmental Chemistry; B. Pina, IDAEA CSIC / Department of Environmental Chemistry; C. Barata, CSIC / Environmental Chemistry

The analysis of lipid disruptive effects in invertebrates is limited by our poor knowledge of the lipid metabolic pathways and of their complete lipoidema. Recent studies showed that tributyltin and juvenoids activated the ecdysteroid, juvenile hormone and retinoic X receptor signalling pathways, and disrupted the dynamics of triacylglycerols in lipid droplets in the crustacean Daphnia magna. This study aimed to explore how ecdysteroids, juvenoids and bisphenol A disrupt the dynamics of phospholipids and neutral glycero lipids in adult daphnias during the reproductive cycle from both lipidomic and transcriptomic points of view. Comparison of the lipidomic profile between treatments and controls revealed relative abundance changes for 194 out of 235 individual lipids detected, corresponding to three classes of neutral glycerolipids (TAGs, DAGs, MAGs) and nine of phospholipids (PCs, LCPs, PEAs, LPEAs, PSSs, LPSs, LPGs, SMSs). Cluster analysis defined two major clusters, one corresponding to control, BPA and 28Es samples, with low levels of TAgS but higher levels of PCs; and another one corresponding to juvenoid-treated samples (PP and MF), with higher levels of TAgS and lower levels of PCs. In addition, subclusters corresponding to lower and higher exposure time were also observed. Transcriptomic analyses identified 1,964 de-regulated genes that were clustered in three groups corresponding to up-regulated gene transcription after 8 or 24h of TBT exposure, and to up- and down-regulated genes after 24 h of exposure to BPA, PC, or TBT. Gene ontology analysis indicated an enrichment of gene signalling pathways involved in lipid metabolism, specifically in lipid catalytic process, triglyceride homeostasis, glycolipid biosynthesis or fatty acid beta-oxidation. This work as supported by the Spanish Research Project EMRISK Code CTM2014-51985-R, (2015-2017). Inmaculada Fuigues acknowledges the Ministry of Economy, Industry and Competitiveness for her fellowship (FPI-MICINN BES-2015-070523).

538 Lipidomics profiling of wild fish to identify patterns associated with pollution exposure
C. Porto, IDAEA-CSIC / Department of Environmental Chemistry; M. Blanco, IDAEA-CSIC; A. Maceda-Beiga, University of Barcelona / Department of Animal Biology

New developments of analytical techniques have allowed the effective identification and characterization of lipids and the development of lipidomics, which has recently emerged as a key technology for human disease research and discovery of biomarkers. However, on an environmental toxicity context, studies are still few, in spite of lipids being considered key molecules for the bioaccumulation of chemicals. This work applies ultra-high performance liquid chromatography coupled to high resolution mass spectrometry (UPLC-HRMS) to characterize the lipidsome of gene signalling pathways involved in fish. This study has been done to the liver of largemouth bass in Lake Apopka, a lake in the Ocala National Forest. We used both a shotgun approach and a targeted approach to quantify perturbations in phospholipids in liver of largemouth bass from Lake Apopka compared to a relatively clean lake in the Ocala National Forest. We used both a shotgun approach and a targeted approach to quantify perturbations in phospholipids in liver of largemouth bass from Lake Apopka compared to a relatively clean lake. Follow up experiments with fish exposed in the laboratory support the changes seen in the field. Cholesterol esters were decreased and cholesterol esters were elevated in the livers of fish from Lake Apopka compared to Wild Cat Lake. This finding corroborates reduced hormone biosynthesis in organochlorine contaminated fish. Other changes in the lipidome are consistent with predicted changes that are related to immune dysfunction. Enrichment in Lake Apopka fish was observed in short chain length free fatty acids, such as palmitic acid and in ceramides, phosphatidic acids and phosphatidylethanolamines. But decreases were observed in sphingomyelins, phosphatidyl-ethanolamines and other phospholipids. These changes are consistent with lipids that are changed due to inflammation and other immune responses. We postulate changes in the lipidome are important biomarkers of OPC contamination.

540 Poster spotlight: WE027, WE028, WE029
Towards a shared understanding of science and risk communication in the context of the inevitability of chemicals and the hazard they may represent (II)

541 Nanotechnology: When shading effects through agglomeration of carbon nanotubes (CNT’s) are confused with toxicity by media and the public - a case example revisited
Dr. Schwab, Adolfo Merkler Institute / Materials Science

Engineered nanomaterials are relatively new contaminants with that can enter the environment via an increasing variety and number of waste streams. The long-term toxicity of nanomaterials is not well understood, and these materials are therefore of emerging public concern. In 2011, we published a press release about our scientific publication on the effects of carbon nanotubes on green algae [1]. We found that the nanoparticles under investigation did not directly affect the algal viability, but indirectly reduced the algal growth via shading and agglomeration. To our surprise,
this press release led to a cascade of secondary articles and events. On the one hand, some online newspapers used our article to produce alarming articles about the dangers of nanoparticles for the environment (example translated from German: “Nanoparticles Identified as Potential Environmental Killers” [2]). On the other hand, some individuals used the press release to draw the oversimplified conclusion that all engineered nanomaterials will eventually agglomerate and therefore be harmless. Nevertheless, most of the media took over the message with no or minor modifications. The press release also triggered surprising responses from within the research institutions. In this presentation, I was invited to briefly summarize the different responses that we got to this press release, and re-iterate the short- and long-term lessons learned from this case study. Most importantly, the ‘real’ work for a scientist writing a press release starts after its publication. News on topics of public concern such as the toxicity of engineered nanomaterials are very closely watched and instrumentalized both by the pro- and the anti-nano community for their respective intentions. Reactions on press releases concerning these topics have to be monitored closely, wrong quotations must be corrected and biased interpretations must be adjusted, in order to provide correct scientific information for the common public. The reward for this work is an overall more balanced communication of the results. [1] Schwab F, Bucheli TD, Lukhele LP, Magrez A, Nowack B, Sigg L, Knauer K. 2011. Are carbon nanotube effects on green algae caused by shading and agglomeration? Environ Sci Technol 45:6136-6144. [3] Georgescu V. 2011. Nanopartikel als potenzielle Umwelthasser ausgemacht. www.lifegen.de/newspaper/shownews.php?getnews=n2011-11-09-3109&pc=02. Accessed 22 Nov 2017 Acknowledgements and Disclaimer - Schwab, F was supported by an Ambizione fellowship of the Swiss National Science Foundation (grant number 168187). Any opinions, findings, conclusions or recommendations expressed in this material are those of the author and do not necessarily reflect the views of the Adolphe Merkle Institute or the SNSF. This work has not been subjected to their review and no official endorsement should be inferred. The author reports no other conflicts of interest and is responsible for the content of the abstract and presentation.

542 Nanotechnology: Communicating scientific findings through media – what could possibly go wrong? Lessons learned from Schwab’s nanotubes G. Obert, UBC / IRES; A. Seal, University of British Columbia / School of Journalism

There is no single effective method for scientists to communicate their findings with the media. Unfortunately, Dr. Fabienne Schwab found this out after publishing a press release about the effects carbon nanotubes (CNTs) have on green algae. When the story hit the press, mainly through an article in Der Spiegel, many readers understood CNTs to be toxic. As a result, Dr. Schwab and her colleagues were accused of fear-mongering. Things escalated to the point that Der Spiegel had to shut down the article’s comments section. Where did things go wrong? How can scientists make sure the media presents their work accurately, but also in a way the general public can understand? We will discuss the diverging roles and realities of science and media, particularly the considerations that scientists and editors need to take into account when they decide to write/publish something.

543 Discussion Nanotechnology

544 Microplastics: The risks of plastics – perceived or real? M. Kotterman, IMARES / Fish

Plastic has not only become a major research topic, it is also broadly covered in popular news. As result the general public knows about the plastic soup and how dangerous it is, supposedly, for wildlife and ultimately for human health. However, the history of plastic research is peculiar. Some of it was straightforward; wildlife choking in plastic does not need much additional proof or QC QA. But, as with many new research topics, the first articles about the dangers of plastic were soon followed by others. The focus was on the presence of small plastics particles even found in seafood, honey and even drinks of different actors related to each story, but also to the society at large, from children to adults, trying to test the changes in those attitudes. The products include videos, cartoons, serious games, interactive tools, specialized courses, etc. Our aim is that if scientists and society have a shared understanding of science and risk communication regarding the problems of the oceans, these can be solved through the individual and collective changes in our attitudes towards the oceans in our daily lives.

546 Ocean Literacy – changing attitudes and behaviour of society in the face of the problems of the oceans

A. Borja, Arts-Technalia / Marine and Coastal Environmental Management

The H2O20 project ResponSEAble (www.responseable.eu) is trying to raise awareness around six key-stories (fishing, eutrophication, renewable energies, coastal tourism, microplastics, and ballast waters), within the four European regional seas. Under the DAPSIWRM framework (Drivers, Activities, Pressures, State, Impact, Wellbeing, Responses, Measures) we are developing products to change attitudes and behaviour of different actors related to each story, but also to the society at large, from children to adults, trying to test the changes in those attitudes. The products include videos, cartoons, serious games, interactive tools, specialized courses, etc. Our aim is that if scientists and society have a shared understanding of science and risk communication regarding the problems of the oceans, these can be solved through the individual and collective changes in our attitudes towards the oceans in our daily lives.

547 Discussion Microplastics

548 General discussion with panel of all speakers about topics emerging from the session

549 Wrap-up and closing

A. Leopold, Calidris Environment BV / Calidris Environment BV; T. Seiler, RWTH Aachen University / Ecosystem Analysis; C. Ajao, ECHA-European Chemicals Agency

Hazard and exposure assessment of chemical mixtures: steps towards increasing the realism of chemical risk assessment (I)

550 Development of a diagnostic toolbox for ecological effects of pollutant mixtures and application to evaluate results from the third Joint Danube survey

A. Focks, Alterra Wageningen University and Research Centre / Environmental Risk Assessment Team; T. Seiler, RWTH Aachen University / Ecosystem Analysis; A. van den Brink, Alterra Wageningen UR / Applied Ecology and Water Quality Management Group b Alterra; B. Deutschmann, RWTH Aachen University
Wildlife is exposed to an infinite number of different combinations of chemicals. Safety and Alternative Methods Unit; S. Bopp, EC Management.

Experimental mixture studies have shown that the toxicity of a mixture is usually greater than the sum of the toxic effects of its individual components. In fact, empirical evidence seems to point to the fact that often substantial mixture effects can occur even though all components in the mixture are present at levels that individually are without observable effects. These observations have lent urgency to the need of evaluating the risks from multiple pollutants both to humans and wildlife. Here, we present a common decision tree and tiered workflow scheme for the assessment of human and ecological risks from pollutant mixtures in European surface waters - case study with > 300 chemicals co-occurring in the Danube. A. Kortenkamp, Brunel University London / Department of Biological and Environmental Sciences; R. Altenburger, UFC Centre for Environmental Research / Institute for Environmental Research Biology V; H. Hollert, RWTH Aachen University / Institute for Environmental Research; S. Kaijser, Faculty of Sciences University of Novi Sad / Department of Biology and Ecology, Laboratory of Ecotoxicology (LECOTOX); T. Backhaus, University of Gothenburg / Department of Biology and Environmental Sciences

Toxic chemicals from point and diffuse sources might impact the ecological status of aquatic ecosystems. Appropriate strategies are needed to identify impacted sites, quantify impacts, or evaluate the causal involvement of chemical contaminants. Since environmental compartments usually contain mixtures of chemicals with low, possibly non-toxic concentrations of the individual compounds, any approach to assess the consequences of toxicological impacts and chemical contamination has to involve concepts for mixture toxicity. However, in addition to toxic chemicals, other non-chemical stressors such as habitat degradation, nutrient pollution, oxygen depletion, pH shifts, hydromorphological changes or others, may also cause a site to fail achieving good ecological status. Since the EU Water Framework Directive (WFD) aims at a good ecological status of all European water bodies through addressing water pollution, for water quality monitoring and assessment under WFD it is necessary to discriminate the impact of such non-chemical stressors from the effects of toxic chemicals. This is challenging, and no single “one size fits all” strategy exists. Therefore, multiparametric approaches, so-called “toolboxes”, are often used. This presentation will show a toolbox for the detection of the ecological impact of chemicals that was developed within the Solutions EU project. It uses a statistically supported, transparent and formalized assessment which could (WOE, etc.) approach. The developed toolbox was applied to the Danube case study, to facilitate evaluation of the very comprehensive data set from Joint Danube Survey 3. The toolbox concept proved to be practical, simple and promising for further studies, with fairly high diagnostic power.

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How can we identify “drivers of mixture risks”? T. Backhaus, University of Gothenburg / Department of Biological and Environmental Sciences; R. Altenburger, UFC Centre for Environmental Research / Institute for Environmental Research UFZ / Effect-Directed Analysis; M. Faust, Faust & Backhaus Environmental Consulting; A. Kortenkamp, Brunel University London / Institute of Environmental, Health, and Societies; L. Posthuma, RIVM / Centre for Sustainability, Environment and Health

Mixtures relevant for human health or the environment can easily contain dozens or even hundreds of chemicals. However, those components do not contribute equally to the toxicity of the mixture. For instance, it is well known that often only a very few chemicals dominate the mixture risk. The European Commission has therefore emphasized in its communication the need to “identify chemical substances that are the main drivers of mixture toxicity”. This could tremendously help to steer future chemical monitoring efforts and risk mitigation measures. However, it is currently unclear how a common definition of the term “driver of mixture toxicity” could be operationalized. In the presentation, we will provide the background of existing approaches to define “mixture risk drivers” and explore the consequences of their application to a real-world case study. In particular, we will discuss: (i) the sensitivity of the various methods to data gaps, (ii) the challenge of tiering the various approaches for risk driver identification, and (iii) the question whether a risk-based ranking is the optimum approach, or whether hazard- or exposure-based methods can be suitable alternatives. In summary, we conclude that, although the identification of drivers of mixture risk would constitute a major step forward to systematize and standardize mixture risk assessment, the concept is currently not yet fit for purpose. Important steps remain to be taken, in order to better conceptualize and operationalize the concept. However, more and more empirical data become available from monitoring studies that provide ample material to explore the applicability of the concept and its consequences for risk management.

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Application of new statistical distribution approaches for mixture risk assessment A. BACKHAUS, T. Faust & Backhaus, Environmental Consulting; S. Hollert, Brunel University London / Institute for Environmental Research Biology V; T. Backhaus, University of Gothenburg / Department of Biology and Environmental Sciences

Wildlife is exposed to an infinite number of chemical exposures under WFD it is necessary to discriminate the impact of such non-chemical stressors from the effects of toxic chemicals. This is challenging, and no single “one size fits all” strategy exists. Therefore, multiparametric approaches, so-called “toolboxes”, are often used. This presentation will show a toolbox for the detection of the ecological impact of chemicals that was developed within the Solutions EU project. It uses a statistically supported, transparent and formalized assessment which could (WOE, etc.) approach. The developed toolbox was applied to the Danube case study, to facilitate evaluation of the very comprehensive data set from Joint Danube Survey 3. The toolbox concept proved to be practical, simple and promising for further studies, with fairly high diagnostic power.

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Towards the development of a framework for applying non-target chemical analysis data within exposure and risk assessment T. Gouin, TG Environmental Research / Safety and Environmental Assurance Centre; R. Parmar, ARN Arnot Research Consulting; J.A. Arnot, ARN Arnot Research & Consulting / Adjunct Professor, Department of Physical & Environmental Science/Department of Pharmacology and Toxicology

There is an increasing trend towards multi-targeted and non-target analysis (NTA) screening methods to increase the number of analytes monitored in biomonitoring and environmental samples. While the possibilities of advances in chemical analytical capabilities have shown substantive development over recent years, application of information related to data reported from NTA represents a challenge to the field of exposure modelling. The case study is based on chemical monitoring data in European rivers, which give realistic empirical data beco

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A common framework for the assessment of human and ecological risks from pollutant mixtures in European surface waters - case study with > 300 chemicals co-occurring in the Danube A. Kortenkamp, Brunel University London / Department of Biology and Environmental Sciences; R. Altenburger, UFC Centre for Environmental Research / Institute for Environmental Research Biology V; H. Hollert, RWTH Aachen University / Institute for Environmental Research; S. Kaijser, Faculty of Sciences University of Novi Sad / Department of Biology and Ecology, Laboratory of Ecotoxicology (LECOTOX); T. Backhaus, University of Gothenburg / Department of Biology and Environmental Sciences

Wildlife is exposed to an infinite number of different combinations of chemicals. There is evidence that single substances that are present below their individual thresholds of effect can still be of concern and contribute to combined effects. A framework for environmental mixture risk assessment (MRA) has been suggested by Backhaus and Faust (2012), however MRA is often hampered by limited data availability. An ecological Threshold for Toxicological Concern (ecoTTC) approach has been recently developed based on a database of more than 100 000 acute and chronic aquatic toxicity data. The tool allows for the calculation of predicted no effect concentrations (PNEC) derived from the underlying data to which an assessment factor (AF) is applied depending on the comprehensiveness of the chemical data and biological database and the chemical distribution, from which the ecoTTC value is derived by calculating the fifth percentile. Other types of chemical toxicity distributions (CTD) are also possible i.e. distribution of acute (LC50) or chronic (NOEC) ecotoxicological data without applying any AF. It is common practice to predict combined effects and risks based on information of the mixture components, most of the time based on the concept of concentration addition (CA). For this case study the sum of risk quotients has been used as a surrogate for CA based predictions. The risk quotient for the mixture (RQmix) is based on the summation of the risk quotients of the individual substances. This approach is conceptually different from CA because the involved PNECs might be based on different groups of species and using different AF. However, it can be used as a screening level approach. If a RQmix>1 is identified, the MRA can be refined by using the sum of toxic units, based on LC50 data. The case study is based on chemical monitoring data in European rivers, which give realistic environmental concentrations and co-exposure scenarios to a relevant number of chemicals. Available ecotoxicological values have been gathered for the identified chemicals from regulatory sources when available, or from the literature and existing database. The possible use of the ecoTTC approach and other type of acute to chronic ratio (CTR) for screening for the appropriate use of NTA data within exposure assessment. Recommendations include a number of suggestions regarding how these data can be better gathered and reported in order to strengthen their applications for chemical exposure and risk assessment, including emerging contaminants.
assumptions about modes of action are introduced. We tested the utility of the scheme by using data on the levels of more than 300 chemicals that occur together in the river Danube, from the Joint Danube Survey 3 (JDS3). For each of the 54 sites along the river Danube we ranked the chemicals in terms of their contribution to a mixture effect, separately for algae, daphnia and fish. We found that the overall mixture toxicity was driven by only approximately 10 chemicals. Substances not yet defined as priority substances under the EU Water Framework Directive made a substantial contribution to combined exposures. We also assessed potential combined risks to humans by evaluating whether water drawn from the Danube would be fit for human consumption. Overall, exposures of concern for humans could not be detected at higher tiers of the assessment. We conclude that the protection goals defined in the Water Framework Directive for freshwater aquatic communities are not achieved for combined exposures at many sites along the Danube.

555 Pesticides do rarely come alone, except in risk assessment - Risk indices of ranked spray series of the project COMBITOX

B. Scholze-Stark, RWTH Aachen University; Gerhard Schuler, Institute for Environmental Research / Institute for Environmental Research; S. Bar, German Federal Environment Agency UBA / Section Plant Protection Products; B. Daniels, RWTH Aachen University / Institute for Environmental Research; T. Frische, Federal Environment Agency UBA / Section Plant Protection Products; M. Liess, UFZ Center for Environmental Research / System-Ecotoxicology; R. Ottermanns, M. Roß-Nickoll, RWTH Aachen University / Institute for Environmental Research; A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics; A. Sybertz, RWTH Aachen University / Institute for Environmental Research; C. Ulrich, German Federal Environment Agency UBA / Section Plant Protection Products; S. Knüllmann, Helmholtz Centre for Environmental Research UFZ / System-Ecotoxicology.

In the European Union, legislation so far strictly regulates plant protection by means of chemical pesticides on the level of single products. Common agricultural practice and manifold pest pressures lead to the exposure of non-target organisms to complex mixtures in a spray series. In the on-going COMBITOX project, we collated a dataset for the actual application patterns of pesticides in 12 representative crops in terms of acreage and pesticide application rates from the years between 2007 and 2015. The data was used for classification of sprayseries by typical mixture patterns, sequences and toxic combinations. Combining all information on empirical use patterns and their regional and temporal variation, we calculated indicators of use intensity and environmental risk. Toxicity exposure ratios (TER) were calculated using standard toxicity data (aquatic & soil organisms) from two publicly available databases: PPDB (Lewis et al. 2016) and ECOTOX (US EPA 2017) and the mere application rates without consideration of exposure pathways.

Only for focal sprayseries, first and higher-tier risk indices (including mandatory risk management measures) for different compartments were computed according to the standard approach as conducted by the UBA within the PPP-authorization procedure in Germany. In sum, 29 risk indices were computed (TER-values). Mixture risk indices were calculated based on the conceptual concentration addition from single-substance TER and summed up per application date. In general, patterns of pesticide use showed that tank mixtures and spray sequences are predominantly in all crops that were considered in our studies. From the data, we revealed that crops group together in classes of use patterns. The cereals received a wide range of different pesticides classes in medium intensities during the whole spraying sequence; vine and apple orchards were dominated by fungicides and high-intensities. Risk exceedances became relevant if multiple individual TER-values (TERsingle) were already close to the critical TER trigger values. Our results emphasize the relevance of the nowadays largely non-regulated tank mixtures for the risk assessment of non-target organisms. In conclusion, we clearly see the necessity to consider realistic exposure assessments of typical treatment regimens as well as effect estimates from appropriate mixture toxicity models in order to describe the “total risk” of the common chemical plant protection practice.

Advances in Soil Ecotoxicology and Risk Assessment of Terrestrial Ecosystems (I)

556 Time-dependent effects of two fungicides and their mixture on enchytraeids and earthworm communities under field conditions

J. Amossé, S. Bart, INRAAgroParisTech; C. PELOSSI, INRA (Institut National de la Recherche Agronomique); M. ROSS, INRAAgroParisTech.

According to the current regulation for the registration of plant protection products on the market, the environmental risk assessment of pesticide use is generally performed under laboratory conditions. Very little information is available in nature, where multiple stresses occur. In this study, we assessed the effects of two commercial formulations of fungicides, i.e., Cupraflor Micro® (composed of 500 g/L epoxiconazole oxycarboxylic acid and 133 g/L dimethoxyrin) and the mixture of both on two groups of terrestrial oligochaetes (Lumbricidae and Enchytraeidae) after 1, 6 and 12 months (i.e., t1, t6 and t12) of exposure under field conditions. We also assessed the feeding activity of soil organisms using the bait lamina method. Our results showed a lower Shannon index for earthworms in the treatment with the mixture of both pesticides (i.e., 1.51 ha-1 of Swing Gold® and 4 kg ha-1 of copper) and in the treatment with the Swing Gold® at ten times the recommended dose (i.e., 15.1 ha-1) after one and six months. We also found a lethal effect of Swing Gold® on anecic earthworms at t1, while an effect of copper on annecic earthworms was observed at t6. Very little data has been observed later at t12. We showed no overall significant difference in total feeding activity, enchytraeid density and diversity between treatments with or without pesticide at different sampling periods. In the Swing Gold® treatment, earthworm community did not recover twelve months after pesticide application. We suggest thus going beyond the ISO norm 11268-3 (2014) for the study of the effects of pollutants on earthworms under field conditions. An unbiased analysis of Oligochaeta community and other functional endpoints (e.g. organic matter decomposition with the tea bag method) over two years to better assess environmental risks of plant protection product use and their mixture. Keywords: Cupraflor Micro®, Swing Gold®, agroecosystems, feeding activity

557 Toxicity of imidacloprid and thiadiazuron towards four Colembolan species C. Lina, Vrije Universiteit Amsterdam / Animal Ecology; C. van Haren, G. Mainardi, Vrije Universiteit Amsterdam / Department of Ecological Science; C. van Gestel, Vrije Universiteit Amsterdam / Ecological Science

Folsomia candida has been used for assessing the toxicity towards non-target soil organisms to pesticides. The mere application rates with two publicly available databases PPDB (Lewis et al. 2016) and ECOTOX (US EPA 2017) and the mere application rates without consideration of exposure pathways.

Only for focal sprayseries, first and higher-tier risk indices (including mandatory risk management measures) for different compartments were computed according to the standard approach as conducted by the UBA within the PPP-authorization procedure in Germany. In sum, 29 risk indices were computed (TER-values). Mixture risk indices were calculated based on the conceptual concentration addition from single-substance TER and summed up per application date. In general, patterns of pesticide use showed that tank mixtures and spray sequences are predominantly in all crops that were considered in our studies. From the data, we revealed that crops group together in classes of use patterns. The cereals received a wide range of different pesticides classes in medium intensities during the whole spraying sequence; vine and apple orchards were dominated by fungicides and high-intensities. Risk exceedances became relevant if multiple individual TER-values (TERsingle) were already close to the critical TER trigger values. Our results emphasize the relevance of the nowadays largely non-regulated tank mixtures for the risk assessment of non-target organisms. In conclusion, we clearly see the necessity to consider realistic exposure assessments of typical treatment regimens as well as effect estimates from appropriate mixture toxicity models in order to describe the “total risk” of the common chemical plant protection practice.
exhibited avoidance behaviour when exposed to acetamiprid or dimethoate. We report the variable effects of 3 insecticides on a range of standard movement behaviours in T. pyri, including distance walked, time moving/not moving, velocity and meandering behaviour. We also report avoidance behaviour measured as the time taken to become trapped in the test arena glue boundary due to attempts to escape the arena. Our results complement existing knowledge of sublethal pesticide effects on NTs by quantifying movement behaviour changes in T. pyri. We are also adding to the knowledge on movement behaviour which is an area of growing interest. We hope to improve the understanding of population-level consequences from changes in movement behaviours caused by pesticide exposure.

559 Should oral exposure in Hypoaspis aculeifer tests be considered in order to keep them in Tier I test battery for ecological risk assessment of PPPs?
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The recent scientific opinion of EFSA addressing the state of the science on risk assessment of plant protection products (PPPs) for non-target arthropods highlighted the need for the inclusion of other relevant exposure routes, besides contaminated soil, in tests from lower tiers. The reproduction test with the predatory mite Hypoaspis aculeifer (OECD 226) is currently being included in the new EU data requirements for the ecological risk assessment (ERA) of PPPs. However, the low sensitivity often shown by this mite towards PPPs, when compared to other invertebrates, makes the test with this species, as it is currently performed, not very useful for Tier I test battery. The current test protocol does not take into account the fact that H. aculeifer is a predatory species, and only considers exposure to contaminants via ingestion; via ingestion, dissolved food, and via contaminated food. Therefore, an adaptation of the test performance, by including exposure via contaminated food, is necessary. The methods described in the standard protocol for mite reproduction test advise feeding the test organisms with fresh preys (e.g. cheese mites Tyrophagus putrescentiae) from uncontaminated breeding containers over the test period but, in a real scenario, this exposure is simultaneous for H. aculeifer and their preys. Thus, through this protocol, the toxicity of contaminants to H. aculeifer might be underestimated. The present study aimed to evaluate the importance of oral exposure to the contaminant as an exposure route to be considered in reproduction tests. Two reproduction tests with H. aculeifer were performed (OECD 226) using artificial soil spiked with increasing concentrations of copper (Cu): 0, 450, 675, 1013, 1519, 2278, 3417 and 5126 mg kg\(^{-1}\). Cheese mites were used as food in both tests but, while in one test cheese mites obtained from normal laboratory breeding cultures (clean preys) were added, in the other test, cheese mites previously exposed to Cu (pre-exposed preys) were used. Results showed that H. aculeifer fed with pre-exposed preys were more sensitive to Cu than mites fed with clean cheese mites. These data support that the route of exposure represents a crucial point in the test performance. Highlands for research in this field are, therefore, to consider the importance of oral exposure in Hypoaspis aculeifer tests and to keep them in Tier I test battery for ecological risk assessment of PPPs. This would be useful for Tier I test battery for ecological risk assessment of PPPs.
related to: methodological framework for S-LCA, goal&scope definition (in particular regarding the system boundary definition), data access, and use of qualitative data, methodologies and selection of indicators for the impact assessment phase. The strengths of the methodology are related to its capability of making the assessment of the product more complete, adding its social aspects to the environmental and economic ones, in addition to the increased transparency and traceability of the supply chain. Along the value chain, the basis of the main challenges for S-LCA identified in this study, recommendations were identified, for further development and implementation of the methodology. The implemented study showed that there is a need for broad improvements of both methodological and communicative issues. To carry out a S-LCA study could be a way to manage social risk thanks to the identification of social hotspots, and to help companies building a targeted strategy for future development of social policies.

563 TBD

564 Social significance analysis of products - considering negative and positive social impacts along the supply chain of leather products
S. Neugebauer, RWTH Aachen University / INAB - Institute for Sustainability in Civil Engineering; M. Traverso, RWTH Aachen

Globally, the leather industry faces complex discussions on social compliance mostly in relation to bad working environments and thus cause severe social impacts for different stakeholder groups along the supply chain due to e.g. unhealthy working conditions. In order to determine social impacts occurring during the leather production processes, social life cycle assessment’s (S-LCA) implementation is of major importance. Practical implementations have to consider indicators and impact categories along the supply chain and should in addidtion provide information on social challenges and chances by means of negative and positive social impacts. When assessing products’ (life) cycles the inclusion of positive social impacts is crucial, as most of the S-LCA indicators can be both positive or negative. Thus, this study aims at providing a social significance analysis (SSA) determining social hotspots along the supply chain including social impacts considering negative as well as positive consequences. Existing social indicators are included, addressing relevant stakeholder groups and impact pathways, e.g. fair wage. New indicators are defined where needed, e.g. to represent the rights for indigenous people affected. The assessments are performed by means of secondary databases, e.g. Social Hotspot Database, and by including primary data gathered at production sites of the European leather producer. The results will provide the challenges and chances of European leather production including the different stakeholder groups affected (e.g. workers) but also positive/negative directions of each social impact category defined (e.g. fair wage as a positive and negative indicator and utility as a purely positive indicator). The SSA is based on the S-LCA of European leather production using both quantitative and qualitative critical topics along the supply chain, e.g. existence of labor laws. Relevant social hotspots are identified. Depending on the indicator direction, social consequences (e.g. benefits in societal health resulting from non-exhausting work hours) can be determined for the stakeholder groups. The inclusion of positive impacts may function as an incentive for improvement and guide the way towards future developments within the European leather industry. The results may also be transferred to further product groups in the global textile and leather industry.

565 Integration of sustainability in industrial research and innovation: perspectives from ArcelorMittal's experience
A. Hettenger, M. Caraty, R. Turconi, ArcelorMittal / Sustainability RD; P. Cortijo, Utopies

The iron and steel industry is both a large contributor to greenhouse gases emissions and a provider of a key material for society's development, being used in a wide range of market sectors such as infrastructure, transport, construction and packaging. Because of its importance and complexity, the development of efficient production processes, minimizing the use of resources, valorization of byproducts, but also to explore the other stages of lifecycle of products that use steel, i.e. to adopt the holistic approach of Life Cycle Assessment. Lifecycle thinking enables ArcelorMittal - the world's largest steel producer - to promote sustainability not only in its own operations but also in the use of its products by customers. For more than a decade, a research team dedicated to sustainability and lifecycle assessment has been supporting the process and product research within the group. However, with more than 1,000 researchers in 12 research centers globally and hundreds of projects carried out every year, it is impossible for a single team to cover systematically the research performed. To this goal, the “Sustainable Innovation Tool” has been developed to enable the researchers of the group to self-assess their projects sustainability. Using the tool, they evaluate environmental and social aspects of their new processes and products and engage in a learning curve for an improved sustainable performance. The presentation will describe the collaborative development of the tool, the different phases of testing and the current start of the deployment across 6 research programs. We will draw on this experience to provide elements that supported its success, pitfalls to avoid. The company is at the start of this journey and seeks a continuous progress, and possible paths for a better integration between our current assessment of industrial research and generic frameworks such as the sustainable development goals will be discussed.

566 Social footprint of a packaging waste deposit-refund system in Spain
J. Muñoz, 2.0-LCA consultants; B. Weidema, Aalborg University; A. Bula, UNESCO Chair in Life Cycle and Climate Change (ESCI/UPF) / UNESCO Chair in Life Cycle and Climate Change. Escola Superior de Comerç Internacional ESCI; F. Pueillan, Universitat Pompeu Fabra UPF / UNESCO Chair in Life Cycle and Climate Change. Escola Superior de Comerç Internacional ESCI

We present a social footprint assessment of implementing a deposit-refund system (DRS) applied to beverage packaging waste in Spain. In a DRS consumers pay a certain amount in concept of “packaging deposit” added to the price of a product and receive the refund back when they return the used packaging. The social footprint developed by Weidema et al. is an example of how the summation of social harm induced by the transfer of money from one societal group to another, and the productivity impact (loss) from missing governance (PG): the difference between the actual purchasing-power corrected value added and the potential value added when all productivity impacts are internalised. The social footprint of an activity can be defined as SF = IR + PL. We compared two scenarios, namely the current situation for household packaging waste in Spain in 2014 (system A), and a hypothetical scenario (system B) where a DRS is implemented. The functional unit was the total amount of packaging waste to be managed annually. Primary data for the two scenarios were obtained from the environmental and economic studies performed as part of this project. Data to quantify the social footprint were obtained from the database Eixiobase v.3.3.10, which was implemented in the software SimaPro. The results show that the social footprint for both systems involves a net social benefit. However system B reduces this benefit by 50% compared to system A. Introducing this DRS system in Spain is expected to lead to a net loss in social benefit compared to the current system. The social benefit of the increased recycling is more than outweighed by the social costs induced by the activities required to achieve these higher recycling rates (collection manually or automatically of packaging waste in shops and supermarkets and the associated transport). This study is an example of how the summation of a social footprint, together with a powerful tool like Eixiobase, can pave the way for an operational approach to social LCA, avoiding excessive data requirements and the long lists of impact indicators currently proposed for bottom-up approaches.

567 Poster spotlight: TH226, TH227, TH228

Developments in the use of bioassays for chemical and environmental risk assessment (I)

568 Application of Equilibrium and Toxicokinetic Models to Understand the Behaviour of Organic Chemicals in In Vitro Toxicity Tests
J.M. Armitage, University of Toronto - Scarborough / Physical and Environmental Sciences; J.A. Arnot, ARC Arnot Research & Consulting / Adjunct Professor, Department of Physical & Environmental Science/Department of Pharmacology and Toxicology

Toxicology testing in the 21st century is expected to rely increasingly on in vitro assays, which now cover a wide range of endpoints including cytotoxicity, receptor binding, protein interactions and DNA binding. In most cases, dose-response relationships from in vitro toxicity tests are reported using nominal concentrations in the test medium despite the known challenges such data introduce for comparing results across different test conditions and between different chemicals and for quantitative in vitro in vivo extrapolation (QIVIVE), Equilibrium partitioning (EQP) and toxicokinetic (TK) models have been proposed in the literature to address some of these issues. The main limitation of equilibrium partitioning models is that instantaneous distribution is assumed and hence uptake kinetics into cells/tissue, cell growth/division and the potential for degradation in the test system cannot be directly included in the calculations. The main objective of this study was to develop a toxicokinetic model (TK) for simulating the behaviour of organic chemicals in in vitro toxicity tests and compare the results to a previously developed EQP model. The toxicokinetic model was applied to a set of hypothetical non-ionic organic chemicals under different scenarios (e.g., biotransformation half-life) and then the results compared with a previously developed in vitro mass balance modeling tool based solely on equilibrium partitioning. We also applied the

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EoQ model to a specific ToxCast assay (ACEA_T47D_80hr_negative assay; cytotoxicity) to illustrate the value of the modelling approaches for QIVIVE and hazard/risk assessment. For relatively persistent chemicals (or in cells/tissue with limited metabolic capacity), the simulated mass distribution using the toxicokinetic model is similar to the equilibrium partitioning model output for test durations greater than 12 h. In such cases, the EoQ modeling approach is deemed suitable for the prediction of water concentrations and the estimation of reliable toxicity metrics for QIVIVE (e.g., membrane concentrations). Of the 306 chemicals included in the ACEA_T47D_80hr_negative assay simulations, approximately 2/3rds had predicted membrane concentrations in the range expected to result in baseline toxicity (membrane dysfunction/necrosis). Chemicals with predicted membrane concentrations well below the baseline toxicity range may act via a specific mode of action and could therefore be prioritized for further investigation.

569 Experimental exposure assessment in in vitro bioassays for organic acids

L. Henneberger, Helmholtz centre for environmental research - UFZ / Cell Toxicology; M. Mühlenbrink, Helmholtz Centre for Environmental Research GmbH - UFZ / Cell Toxicology; B. Fischer, Helmholtz Centre for Environmental Research GmbH - UFZ; B. Escher, Helmholtz Centre for Environmental Research GmbH - UFZ / Cell Toxicology

Improved assessment of exposure in in vitro toxicity assays is essential for the application of in vitro data for chemical risk assessment. Equilibrium mass balance models have been developed to convert the reported nominal effect concentrations of chemicals to static exposure concentrations. In order to establish a methodological foundation for the determination of static effect concentrations, we used two-dimensional plate cultures of unicellular microalgae to determine system level equilibria. Kinetic models were developed for the different cellular matrices like cell culture media and cell suspensions on the one hand, but also for direct measurement of exposure (i.e., Cₕ). Because polymers like polydimethylsiloxane that are typically used for solid phase extraction (SPME) are not suitable for charged chemicals, C₁₈-coated SPME fibres were used in this study, that have been previously reported to have high sorption capacities for charged chemicals. Eight organic acids were chosen for the experiments: diclofenac, 2,4-D, ibuprofen, naproxen, toleramide, warfarin, tricosan, and gentamicin. These compounds were all found to reach equilibrium and water was established within 4 h and the determined fibre-water distribution ratios were reproducible (SD ≤ 0.1 log units). Because the sorption of some of the chemicals to the fibres was concentration dependent, it was required to calibrate the fibres for the desired concentration range. The SPME method was applied to measure Cₕ in cell culture media. At low chemical concentrations the results from the binding experiments gave the predictions from mass balance modelling approach. However, saturation of the medium was observed at high chemical concentrations and further experiments will be necessary to investigate for which chemicals and at which concentration levels saturation occurs and if it is required to incorporate non-linear binding into existing exposure models for in vitro bioassays.

570 A versatile and low-cost open source pipetting robot for automation of toxicological and ecotoxicological bioassays

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The contrasting demands of performing bioassays in compliance with regulatory, ethical and logistical requirements are usually met by calling for automation technology to assist with automated handling and analysis of multiwell plates. Such systems are typically highly sophisticated and thus costly. As a consequence, the availability of pipetting robots, liquid handlers, and stacking units in environmental monitoring is generally scarce. As a potential solution, we developed a simple and low-cost, versatile open-source pipetting robot that has a small footprint. The construction of the pipetting robot was realized mostly using readily available parts, and partly using open-source hardware. We tested its precision in automated 2-fold dilution series and used it for exposure of zebrafish embryos (Danio rerio) – a common model species in ecotoxicology - to cadmium (Cd) and permethrin. As expected, concentrations of permethrin rapidly decreased after initiation of static exposures and after each renewal in the semi-static exposure experiments. No such drastic differences were observed for exposures conducted using the pipetting robot. The accuracy of the pipetting steps was generally high. The apparent toxicity was not only greater in zebrafish embryos exposed to permethrin and cadmium using manual semi-static renewal (24 h interval) than in the automated semi-static exposure using the pipetting robot (1 h interval). Thus, we were able to confirm that any attempt to keep exposure concentrations as constant as possible will yield more realistic assessments of toxicity. In this respect, exposure using our pipetting robot can be hypothesized to be similar to flow-through exposure, which is, however, typically more labor- and cost-intensive. With minor modifications, this versatile system can be used in a variety of different setups and environments. Because its construction and operation are very cost-effective and do not require any specialized personnel, provisioning of instructions to replicate this design has makes automation technology accessible to a much higher number of laboratories around the world.

571 An intestinal fish cell barrier model to assess absorption of poorly soluble organic chemicals in vitro

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Permeation of organic chemicals from the aquatic environment across cellular barriers is a critical step for accumulation in organisms, such as fish. To better understand the underlying processes, we aim to study the role of the fish intestine as a barrier for hydrophobic and volatile chemicals. The function of the intestine in these processes is experimentally impractical to assess on a routine basis. Additionally, hydrophobic and volatile chemicals are difficult to work with, due to their low water solubility and high volatility. Therefore, we here combine a recently developed in vitro epithelial barrier model using the rainbow trout (Oncorhynchus mykiss) intestinal cell line RTgutGC, and a newly constructed chamber that enables stable chemical exposure concentrations. In this setup, we measured the permeation of 10 fragrance compounds with a range of volatility (logHLC = 5.8 to -2.2) and hydrophobicity (logKOW = 3.6 to 5.7). The RTgutGC monolayer partly presented a physical barrier for the permeation restricting the fragrance transfer from the apical to the basolateral compartment. The calculated permeation rates across the cell layer combined diffusion controlled permeation and intestinal biotransformation. The involvement of biotransformation within the cell monolayer was further supported by experiments at 4°C and the measurement of cell associated chemical concentration. We determined the chemical distribution in all different compartments of this model, which correlated with the logKOW. The chamber enabled stable exposure concentrations and close to full recovery at the exposure medium over the entire measurement time. We believe that this barrier model can help to develop strategies to link in vitro permeation with the prediction of bioaccumulation factors for fish. Moreover, the well-defined exposure scenario in our system comprises a prerequisite for studying mechanisms underlying chemical permeation, such as active transport or biotransformation.

572 A new paradigm in water sampling - how can we challenge the needs of effect-based monitoring?

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In vivo and in vitro bioassays (effect-based methods, EBM) are increasingly used for the water quality monitoring to complement chemical analysis. In a holistic point of view, sampling is the starting point and an integrated part of the whole analysis workflow. However, sampling for effect analysis is more challenging than for chemical analysis. Thus, the aim of this paper is to discuss (1) the requirements and challenges of sampling for EBM and (2) to present the recently developed large-volume solid phase extraction approach and apparatus (LVSPEx) as a
promising technology to overcome the disadvantages of traditional sampling techniques with respect to EBM. The first challenge is the demand of water to be enriched which is dependent from the number and extracts consumption of the bioassays used for the assessment. The second challenge is the recovery and carry-over of the potential toxicity from the water sample to the vessels or wells of the bioassay. The third challenge is the representativeness of the sample. The successful implementation of EBM strategies requires the availability of automated sampling devices which allow the sampling of larger water volumes, guarantee the sampling integrity and makes it possible to take representative samples over a longer period or during events such as heavy-rain- and flood-events. A solution to overcome the disadvantages of classical sampling methods and devices is the recently released LVSPE approach and apparatus. It brings the SPE onshore, allows full automation of processing and avoids the transport of larger water volumes to laboratory for filtration and extraction. LVSPE was comprehensively assessed with respect to recovery and carry-over of effects. It has been shown that LVSPE is applicable in monitoring and survey programs, to assess surface water and wastewater with effect-based tools and to unravel one of the causes of mutagenicity in the river Rhine using effect-directed analysis. Thus, LVSPE is a promising technology for the implementation of EBm for water quality monitoring in European and worldwide water quality monitoring. LVSPE is commercially available from MAXX GmbH. However, we declare no conflict of interests as all results presented are scientifically justified.

573 Prioritization of non-target screening suspects based on semi-quantitative concentrations and ToxCast in vitro toxicity data

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In addition to target analyses of chemicals in water samples, non-target analyses are increasingly being applied. The aim of this study was to develop an innovative prioritization tool for chemicals of emerging concern for drinking water, by combining HRMS data with high throughput toxicity data from EPA’s ToxCast database. To increase the health relevance of the prioritization method, both semi-quantitative concentrations (internal standard equivalents) in the water samples (as a measure of exposure) and toxicity classes based on 5th percentile AC₅₀ values (as a measure of hazard) were included as these form the basis for health risk assessment. A procedure to collect chemical-specific toxicity data from the ToxCast database and a scoring methodology for detected suspects were developed and applied to different types of water samples (sewage treatment plant effluent, surface water, ground water and drinking water) to prioritize suspects for identification and further risk assessment. ToxCast data were collected from the EPA’s online ToxCast database repository. Assay endpoint AC₅₀ values (the concentration at which 50% of the maximum response is achieved) were extracted from the ToxCast database for the tested chemicals (suspects). All ToxCast assays were included in this hypothesis-free analysis. To reduce the impact of very sensitive assay endpoints, the 5th percentile of the range of AC₅₀ values of a suspect chemical in ToxCast assays was used as a measure of its toxicity. More than 7000 structures were detected in these water samples by HRMS non-target screening analyses, and these could be linked to >1000 suspects from a curated suspect list of >5000 EU and water relevant chemicals. The ToxCast database contains in vitro effect data for 549 of the 1073 suspects present in the water samples. Many suspects were prioritized based on toxicity and semi-quantitative exposure levels that were not prioritized earlier based on exceedance of the Threshold of Toxicological Concern. After confirmation of their identity, the prioritized suspects are candidates for a in-depth risk assessment based on all available toxicity data, for introduction in monitoring programs or for further risk management measures. Standardization of prioritization schemes for suspect screening approaches may be needed for further introduction of these techniques in water quality regulations. Funded by the Joint Research Program of the Dutch water companies (BTO, project 400554-214).

From detection to action: advancements in assessing and managing highly fluorinated compounds

574 Toward the Comprehensive Profiling of Zwitterionic, Cationic, and Anionic Perfluoroalkyl and Polyfluoroalkyl Substances in Firefighting Foam Impacted Soils

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In recent years, the comprehensive analysis of both aqueous film forming foam (AFFF) formulations and environmental samples aided to reveal the identities of novel classes of perfluoroalkyl and polyfluoroalkyl substances (PFASs). Following the prioritization of firefighting activities or fire emergencies, it was observed that the soil is typically the first environmental compartment impacted. In such samples, newly identified PFASs could surpass the concentrations of legacy PFASs by orders of magnitude, indicating the need for in-depth characterization of their transport potential and effects in ecosystems. The methods currently available for the analysis of perfluoralkyl acids (such as PFOS or PFOA) could, however, seriously underperform for certain newly-identified PFASs. Severe discrepancies were noted as regards the extraction efficiency of cationic and zwitterionic PFASs between soils of variable textural class and organic matter (OM) content, which could be compensated through development of concentration specific extraction mechanisms. Given the aforementioned limitations, the present study set out to propose a suitable preparation procedure for the multi-residue analysis of PFASs in AFFF-impacted soils. A total of 89 PFASs, representing >20 distinct chemical classes previously discovered in AFFF formulations or across AFFF-impacted sites, was therefore evaluated. Multiple extraction methods were assayed to recover PFASs from AFFF-impacted soils and 28 industrial textiles have been investigated in-house with AFFFs and aged. The optimized method presented quantitative or near-quantitative PFAS recoveries from diverse soils and limited matrix effects were noted. The method was applied to a limited survey of firefighting training areas in eastern Canada, showing the prevalence of betaine-based (e.g., 6,2 FTAB, 9:1:2 FTB) and amine-based (e.g., PFHxS,PFAS) PFASs at such sites.

575 Investigation of perfluoralkyl and polyfluoroalkyl substances in products used for building industry as well as industrial textiles and their possible contribution to water contamination

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Perfluoropolyalkyl and polyfluoroalkyl substances (PFASs) have been widely studied in environment, outdoor clothes and fire fighting foams. As a consequence of the hazardous environmental properties of some PFASs, such as persistence, bioaccumulation and toxicity, their fate has been widely discussed. Due to their water, dirt and moisture repelling properties, PFASs are suitable for a variety of applications and possess a lot of possible entry pathways that were identified in prior studies. Although, advertisement and material safety data sheets indicate a rather extensive use of PFASs in building materials and industrial textiles, only few studies deals with investigation of these materials. A total of 23 samples from products used in building industry and 28 industrial textiles have been investigated in the course of this project. Monitoring covered 29 PFASs with a chain length of C4 to C14, including carboxylic acids, sulfonic acids, sulfonamides and fluorotelomer alcohols. PFASs of diverse chain length (C4-C14) were detected in 31 of 51 investigated samples. Concentrations of perfluoroalkyl acids were up to 430 µg/kg for highly contaminated samples. FTOHs were even detected in concentrations up to several mg/kg. However, FTOHs need to be further investigated since they may be false positives resulting from the low selectivity of utilized transition for FTOH quantification. In addition to performed investigations, rinse of samples of e.g. buildings should be performed to prove suggested entry pathways. Furthermore, additional method development has to be performed in order to better illustrate entry pathways.

576 The growing role of seafood consumption for exposures to legacy PFASs: Evidence in Longitudinal Birth Cohorts from the Faroe Islands

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Rapid declines in legacy poly- and perfluoralkyl substances (PFASs) have been reported in human populations globally following changes in production since 2000. However, temporal shifts in exposure sources are not well understood and thus, information for mitigating future exposure pathways of PFASs are needed. The concentrations of 19 PFASs (SPFASs) measured in children between 1993 to 2012 from a North Atlantic fishing community (Faroe Islands) where pilot whale is part of the traditional diet. Median SPFAS concentrations in children (ages 5 to 13 years) peaked in 2000 (47.7 ng mL⁻¹) and declined significantly by 14.4% yr⁻¹ to 8.7 ng mL⁻¹ in 2012. Perfluorocarboxylic acids (PFCAs) with nine or more carbons (C≥9) were strongly associated with mercury in children’s hair, a well-established proxy for seafood consumption, especially perfluoroundecanoic acid (PFUnDA, z = 0.72). Toxikokinetic modeling revealed PFAS exposures from seafood have become increasingly important (53% of perfluorooctane sulfonate: PFOS in 2012), despite a decline in whale consumption in recent years. A previous study reports PFASs in Faroese drinking water were below detection. We thus infer that even in a major seafood consuming population, guidelines in SPFASs exposure after 2000 were achieved by the rapid phase out of PFOS and its precursors in consumer products.
Membrane-water partition coefficients to aid PFAS risk assessment.

S. Droge, University of Amsterdam/IBED Institute / IBED

Widely varying structures of fluorinated compounds have been detected ubiquitously in humans and the environment. Due to the limited understanding of basic physical-chemical properties of any of these PFASs, risk assessment (RA) models still provide highly uncertain outcomes. Most PFASs are ionogenic, and act as surfactants. As a result, octanol-water partition coefficients (Kow) cannot be determined experimentally. Due to the lack of experimental data, QSARs to predict Kow are not properly calibrated for any perfluorinated ionogenic compounds. Furthermore, the dissociation constant (pKd) of PFASs has proven to be difficult to determine experimentally and is simply unknown for most emerging alternative PFASs. This may lead to high uncertainty on the fraction of ionized and neutral species at a certain environmental pH of emerging PFASs, and the link to the chemical’s “hydrophobicity”. One of the main applications of a Kow value in RA models is to relate a chemical’s “hydrophobicity” to bioaccumulation and toxicity. It is therefore utmost surprising that hardly any data is available on measurements of sorption data to (phospho)lipid, for which assays are readily available and that lack all the concerns about experiments with octanol. Phospholipids are in general the key tissue component to sorbs ionogenic surfactants, and relate directly to baseline toxicity levels and bioaccumulation. The current study evaluates the results of two experimental tools to measure sorption of standard PFAS structures to artificial phospholipids: retention on immobilized phospholipid chromatographic column and solid supported lipid membranes. The current study also evaluated to what extent quantum-chemical software COSMOtherm, which does not require experimental data, can be used to predict Kow simply by taking 3D molecular charge densities into account, is able to predict for emerging PFASs both the membrane-water partitioning (Kow) of the ionic perfluor species, and the predictions on pKd. Whereas COSMOtherm accurately predicts Kow for cationic hydrocarbon surfactants, it strongly misinterprets the membrane affinity of anionic perfluorinated surfactants. It does show promising predictions on pKd of alternative PFASs e.g. GenX and PFAS sulfonamide (PFOSA) to be a strong acid, whereas recent studies and reports suggested PFOSA to be a neutral PFAS.

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Impacts of ocean circulation on the marine PFOS burden in an era of geographically shifting emissions

C. Wagner, Harvard University / Harvard John A Paulson School of Engineering and Applied Sciences; C. Thackray, Harvard University / School of Engineering and Applied Sciences; X. Zhang, Wisconsin Department of Natural Resources / St. Eustatius and Saba Office of Pollution Control and Resource Reuse, School of the Environment; E.M. Vandermeulen, Harvard University / School of Engineering and Applied Sciences

Concerns over the persistence, bioaccumulation potential and toxicity in organisms prompted the inclusion of perfluorooctane sulfonate (PFOS) in the Stockholm Convention in 2009. The ocean is thought to be the terminal sink for most PFOS residues, but simply requiring takes 3D-molecular charge densities into account, is able to predict for emerging PFASs both the membrane-water partitioning (Kow) of the ionic perfluor species, and the predictions on pKd. Whereas COSMOtherm accurately predicts Kow for cationic hydrocarbon surfactants, it strongly misinterprets the membrane affinity of anionic perfluorinated surfactants. It does show promising predictions on pKd of alternative PFASs e.g. GenX and PFAS sulfonamide (PFOSA) to be a strong acid, whereas recent studies and reports suggested PFOSA to be a neutral PFAS.

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PFAS pollution at airport sites: point and diffuse sources, fate and transport and remediation

S. Hille, G.D. Breivik, Norwegian Geotechnical Institute

Using Norwegian airports and fjords as case studies this work will present the identification of point and diffuse source PFAS inputs, the environmental behavior (partitioning) of these compounds in relation to their presence in soil, ground water, surface water and biota as well as suitable remediation methods for different sites. The once highly desirable physicochemical properties of PFAS (resistance to heat, water and oil) has led to a large-scale environmental problem as these properties go hand in hand with a low degradative potential, high persistence, high mobility and toxicity. At airport firefighting training facilities aequous firefighting foams (AFF) containing PFAS have been used in order to practice extinguishing fires. This has led to a point source input of pollution of soils, waters and biota in close proximity to these areas. The use of PFAS containing substances by the textile and paper making industries, as well as their presence in waste sent to landfills represent several diffuse source pollution pathways. The case studies presented encompass all of these inputs, and methods used to monitor the behavior of PFAS in soil, ground water, surface water and biota (including the use of passive samplers) will be presented. Understanding the partitioning and leaching behaviors of emerging alternative PFAS compounds allows more informed regulatory decision making and given that the regulation of PFASs is currently under the spot light this is of great importance. Perfluorohexane sulfonate were recently included on the list of Substances of Very High Concern in REACH. PFOS is included in the water framework directive and an environmental quality standard is often used in order to set clean up targets. The remediation of PFAS impacted sites presents unique challenges and current remediation of water often relies on pump and treat using activated carbon filters to sorb PFAS. There are fewer suitable remediation methods for soils. Through the case studies presented, different remediation methods that are currently being used in the field and lab (pump and treat and sorbent amendment) will be presented. Sorbent amendment has been shown to be a promising approach with reductions of PFAS leaching up to 99%.

Improvements in environmental exposure assessment: State Key Laboratory of Pollution Control and Resource Reuse, School of the Environment and Applied Sciences; C. Thac 2019

G.D. Breedveld

- water partition coefficients to aid PFAS risk assessment.

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entrenched in a grease base, it is proposed that a lack of exposure based on low solubilities and/or bioaccessibility is taken into consideration when registering the substances under REACH. This is a pragmatic approach for a group of substances that have low hazard potential and avoids conducting unnecessary vertebrate animal testing. The ERGTC strategy for registering grease thickeners under REACH, taking into consideration bioaccessibility, will be presented, including proposed “limits for leaching”. This approach could be expanded to include other types of similar substances which occur in situ in an inert carrier such as base oil.

584 The durability criteria: a pragmatic and sound approach to the exposure assessment of nano-enabled agrochemicals
M. Others; University of York / Department of Environmental Geosciences; R.S. Kookana, CSIRO / Land and Water
After many years of research and development, nano-enabled agrochemicals are starting to make their way into the market. Evaluating nano-enabled agrochemicals against conventional analogues is essential to assess the new risks and benefits associated with the technology, and this raises a number of issues for regulators. The ecological risk assessment of nano-enabled agrochemicals is likely to differ from that of conventional products and new parameters are needed to allow an adequate evaluation of the new products. The majority of products currently in development consists in nanocarrier systems loaded with a registered AI. For this type of products, a priority for assessment is to establish the time during which the AI remains associated with the nanocarrier, i.e. the “durability” of the AI-nanocarrier complex (1). Koopman et al. (2) presented a series of guiding principles for the regulatory evaluation of environmental risk associated with nano-enabled pesticides, including a conceptual strategy relying on the durability parameter. A group drawn from regulatory agencies, academia, research, and the agrochemicals industry recently offered a perspective on relevant considerations pertaining to the problem formulation phase (3). A case study (pendimethalin–aqueous suspension formulation) was chosen to test and the framework proposed during exposure assessment. The scheme serves as a useful base to guide additional requirement and help regulators to take informed decisions without having to systematically perform a comprehensive new nano-specific assessment. One of the key conclusions is that an early and reliable measure of the durability of the AI-nanocarrier complex under relevant conditions is key to the assessment of nano-enabled pesticides. There is currently no standard protocols to measure the durability of the AI-nanocarrier complex and robust methods for its measurement are urgently needed. References: 1. Kah M, Hofmann T. 2014. Environ. Int. 63:224–235. 2. Kookana RS et al. 2014. J. Agric. Food Chem. 62:4227–4240. 3. Walker GW et al. 2017. J. Agric. Food Chem. doi:10.1021/acs.jafc.7b02373.

585 Can we model emissions, fate and exposure on a global scale? A case study of PCB 153 in human milk
M. McLachlan, Stockholm University / Environmental Science and Analytical Chemistry (ACES); E. Undeman, Stockholm University / Baltic Sea Centre; F. Kookana, CSIRO / Land and Water
The modeled co-occurrence of PCB 153 and other PCBs in human milk samples can provide insights into exposure pathways. An exposure assessment using the System for Predicting Emissions and Chemical Reactions (SPERC) was developed, by grouping similar uses, mainly based upon their physical form and application mode. Next, a Fertilizers Environmental Exposure tool (FEE) tool was developed, since in the standard REACH models for environmental exposure assessment (EUSES, ECETOC TRA, CHESAR), no local scenarios were developed for direct field applications. PCBs on agricultural fields to surface water are not taken into account. In addition, important output pathways, for example via crop uptake and harvest, are largely not considered in these tools. Quantitative exposure scenarios, resulting in the calculation of realistic worst-case local Predicted Environmental Concentrations (PECloc) for fertilizers in the various environmental compartments (soil, water, sediment) were established. As the main focus of the FEE tool is on micronutrients such as manganese, copper and zinc, which are identified as hazardous for the aquatic environment; the tool allows for assessment of other inorganic and organic substances in fertilizers as well. Conceptually, the tool has been based upon existing REACH exposure modelling, but is adapted for fertilizer use and the assessment of substances downstream from other chemical industries. In order to improve harmonization and communication within the supply chain, generic exposure scenarios have been developed for a number of micronutrient and SPERC combinations. Collectively, the development of SPERCs, the fertilizers environmental exposure tool and generic exposure scenarios, allow for a systematic review of environmental exposure assessments of fertilizers and pesticides under REACH regulation. Further information on the project, including downloads of the FEE tool and SPERC factsheets can be found via www.fertilizersEurope.com.

583 Bioaccessibility of grease thickeners and the implications for REACH regulation
R.J. Brown, wca consulting; R. Smith, wca; P. Whitehead, wca consulting; J. Dawick, G. Whale, Shell Health / Risk Science Team; A. Dodos, Eldorns; T. Halmans, Shell Global Solutions International / Analytical Department
An intrinsic component of greases are the grease thickeners which include a diverse range of substances including metal soaps, metal-complex soaps and polyoxylates. These different thickeners impart different technical properties on the final grease. Although individually registered under REACH as isolated substances (soil, water, sediment) were established. The human diet in ACC countries such as Australia, New Zealand, Canada and the USA has been shown to be highly correlated, with a correlation coefficient of 0.76. For 49 out of 78 data points, the predictions and observations agreed within a factor of 4. The model over-predicted the concentrations in central Europe and under-predicted the concentrations in much of Africa, in particular West Africa. Potential weaknesses identified in the chain of models include an under-prediction of the rate of decrease in PCB concentrations in air since the 1980s and inadequate treatment of food sourcing. We conclude that we have come a long way towards meeting this grand challenge for PCB 153, but there remains room for improvement! References U. Schenker et al. Environ. Sci. Technol., 2005, 39, 8343-8441. K. Breivik et al., Environ. Sci. Technol., 2016, 50, 798-805. M. MacLeod et al., Environ. Pollut., 2011, 159, 1442–1445. G. Czub and M.S. McLachlan, Environ. Toxicol. Chem., 2004, 23, 2356–2366.

https://undatacatalog.org/dataset/gemsfood-consumption-database M. van den
Risk assessment of Nanomaterials: innovative approaches and application of recent research developments to regulatory science

586 Regulatory tools and activities for environmental risk assessment of nanomaterials in ECHA

ECHA implements the REACH Regulation (EC No 1907/2006 (Registration, Evaluation and Restriction of Chemicals), the Biocidal Products Regulation (BPR, Regulation (EU) 528/2012)), and the Classification, Labelling and Packaging (CLP) Regulation (EC No 1272/2008). Industry and authorities need to fulfill their obligations regarding these regulations also for nanomaterials as for any other form of a substance. Nanomaterials are implicitly covered by the substance definition of REACH Regulation 1907/2006 although there are no explicit requirement laying down NM specific obligations. ECHA’s experience has shown that REACH would benefit from nano-specific provisions. The BPR has partly implemented the Commission recommendation of 18 October 2011 on the definition of nanomaterials article 3(1)(xi). It states that the approval of an active substance does not cover nanomaterials explicitly mentioned (Article 6). ECHA currently performs three type of activities to implement REACH, CLP and BPR regulations and to support these processes aiming at ensuring safe use of nanomaterials (NM): REGULATE: formal processes under regulatory frameworks, whereby ECHA uses the legal instruments available under REACH (dossier evaluation, authorisation and restriction), CLP and BPR; SUPPORT: helpdesk, meetings with stakeholders and with Registrants, Nanomaterials Expert Group (NMEG); COMMUNICATE: ECHA Nanomaterials web-site, conferences, workshops, communication throughout the supply chain and in a broader context e.g. EUON and press. This presentation will provide a summary of the multiple actions taken by ECHA to address NM under REACH, CLP and BPR: Dossier and substance evaluation, NMEG, EUON and ECHA’s involvement at OECD level. Communication is currently considering modifying some of the technical provisions in the REACH Annexes. This would allow more efficient efforts towards safe use of NM and decreased uncertainties in the regulatory processes. In addition, ECHA highlights the need for good coverage of standard methods applicable to NM to produce adequate information for regulatory risk assessment.

587 Building a Risk Assessment Framework for Nanomaterials in Canada
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Despite the potential benefits associated with the use of nanomaterials, concerns also exist as to potential environmental and human health risks as a result of environmental exposure to nanomaterials. Canada regulates chemical substances, including nanomaterials, under various regulatory mechanisms. The Canadian Environmental Protection Act, 1999 (CEPA) and the Chemical Management Plan (CMP) are key in safeguarding Canadians and the Canadian environment from potentially harmful substances. Under the CMP, the current activities to address nanomaterials include identifying data needs, developing tailored strategies and approaches, work planning and strengthening collaboration with national and international partners and stakeholders. To support these efforts, Canada is developing a science based risk assessment framework (RAF) to guide legislative and regulatory risk assessments of nanomaterials under CEPA. This science based RAF will inevitably touch on many issues faced by other countries and regulators. Our communication strategy includes consultation and collaboration with multidisciplinary experts, the public and stakeholders. Online collaboration tools will be used to encourage participation. A national expert panel will be consulted in the spring 2018, and SETAC Europe would be the first attempt to reach out to the international scientific community for support. We hope to use this venue to generate new partnerships and to strengthen existing ones. Critical questions based on the challenges identified by Canada over much more than a decade of regulating new nanomaterials, and by the international scientific community looking at risk assessment challenges for nanomaterials, will be highlighted in this presentation.

588 Inventory of available tools, methods, approaches and best practices on nanomaterials/nanotechnologies
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The EC4SafeNano initiative, founded by Horizon 2020 in 2010 is an ongoing effort to build a European Centre for Risk Management and Safe Innovation in Nanomaterials and Nanotechnologies. EC4SafeNano aims to bridge the gap between scientific knowledge on hazard and risk, and ‘fit-for-purpose’ risk management tools and strategies supported by measurement and control methods. The Centre of European organisations will offer services for Risk Management and Safe Innovation for Nanomaterials & Nanotechnologies. One of the first actions of the project was developed an inventory of available resources related to nanosafety issues. Indeed, until now, many tools and studies which aim to improve human and environmental nanosafety have been developed by research organisations and EU funded projects. Therefore this inventory have the aim to give a useful overview on tools, methods, standards, standard operating procedures (SOPs), guidance documents and best practices in nanosafety. Quality criteria are included to give users the possibility to select or sort based on examples, for instance classification that approved the standard (and thereby indirectly the procedures followed to come to a standard), the level of evaluation and validation of the resources or the acceptance of the resource in view of the REACH legislation. During the project and after the duration of the project this overview will be updated when new information or updated versions of resources becomes available or when new resources are introduced by a mechanism later to be defined. Moreover this inventory will be published on the EC4SafeNano website. There is a large number of Standards (77) and SOPs (136) that can help the end user to conduct testing on toxicity and eco-toxicity, or measurements in workplaces and environment. One important issue is the scarce number of training available only 5. This aspect will be addressed in the EC4SafeNano project to identify the specific training needs and that can be useful to promote the understanding of environmental risks and the amount of information related to the environmental assessment of nanomaterials and nanotechnology is very low. This aspect is covered only in 6 out of the 28 included tools, and 4 out of 43 Guidance and Best practices. This shows that, at the moment, aspects such as eco-toxicity and methodologies for the environmental risk assessment and life cycle assessment have most likely not been deeply investigated.

589 The Application of Ecotoxicological Tools to Safer-by-Design Strategies for Engineered Nanomaterials
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OECD test guidelines using aquatic organisms have been identified as suitable starting points for identifying risk that engineered nanomaterials (ENMs) risk represent for the environment. However, standardized testing (mainly relevant for freshwater species) may not adequately capture the risk and dynamic behavior of ENMs in the environment, in particular for aquatic regions like coastal areas which are the ultimate sink for all land based contaminants. As a result, the addition of less conventional organisms to regulatory projects can promote the characterization of environmental risk nanomaterials pose. Mytilius species have a long history of being used as sentinel organisms to characterize ecosystem health and can be useful for the understanding of environmental risks as a result of emerging contaminants like ENMs pose. This information can be particularly useful for the development of safer-by-design strategies as implementation in risk characterization can promote the identification of which products an industry creates pose the lowest environmental risk. In this context, under project the Horizon 2020 project NanoReg2, the aim of this study was to demonstrate the implementation of a safer-by-design strategy for ENM development. Three carbon nanofibers (CNFs) were provided by an industrial partner in order to characterize the potential environmental risk their products pose. The aim of the study was to compare the original product (GAFN) to a new scaled up production process for the CNF (GATam) as well a graphitized version of the product (GANf). The study implemented regulatory testing using Daphnia magna and Pseudokirchneriella subcapitata as well as hemocyes from the marine mussel Mytilus edulis (M. edulis) following in vitro and in vivo testing on subcellular endpoints. The testing strategy was conducted in order to demonstrate the suitability of both regulatory testing (OECD tests) as well an in vitro screening strategy on M. edulis hemocytes to characterize the environmental risk posed by ENMs in the context of safer-by-design and its application to industry. In addition, recommendation and discussion on protocols used to test this CNF are provided.

590 Minimising the risk posed by TiO2 nanomaterials used in sunscreen throughout the entire product lifecycle
nanomaterials contained in the sunscreen can be released into rivers, lakes, sea shores, and/or sewage treatment plants. Their fate and impact in these different systems is largely determined by the surface properties, i.e. the coating type and lifetime. This project aims to develop the eco-design of sunscreens through the minimization of risks associated with nanomaterials incorporated into the formulation. All stages of the cream life cycle must be considered in this light, from its manufacture to its end of life, through its use by the consumer and its impact on the aquatic environment. By considering each development stage of the sunscreen, from the choice of UV-blocker and its integration into a cosmetic formulation, to the knowledge of the risk involved in this choice all along the product lifecycle, an eco-design approach can be achieved and risk can be minimized. The present work combines industrial companies specialising in cosmetic formulation with academic research experts in the fields of exposure, toxicity and lifecycle assessment. Sunscreen fabrication, risk for the consumer by dermal exposure, risk for the direct aquatic environment and risk related to the end of life of the product are as many key steps of the sunscreen lifecycle that were investigated in this project.

591 Environmental risk assessment of engineered nono-SiO2, nano iron oxides, nano-CeO2, nano-Al2O3, and quantum dots
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Many research studies have aimed to investigate the ecotoxicological hazards of engineered nanomaterials (ENMs). However, little is known regarding the actual environmental risks of ENMs, combining both hazard and exposure data. The aim of this study is to quantify the environmental risks for nano-Al2O3, nano-SiO2, nano iron oxides, nano-CeO2, and quantum dots by comparing the predicted environmental concentrations (PEC) with the predicted no effect concentrations (PNEC). The PEC values of these five ENMs in fresh waters in 2020 for northern Europe and southeastern Europe were taken from a published dynamic probabilistic material flow analysis model. PNEC values were calculated using probabilistic species sensitivity distribution (PSSD). The order of the PNEC values was quantum dots < nano-CeO2 < nano iron oxides < nano-Al2O3 < nano-SiO2. The risks posed by these five ENMs were demonstrated to be in the reverse order: nano-Al2O3 < nano-SiO2 < nano iron oxides > nano-CeO2 > quantum dots. However, all risk characterization values are four to eight orders of magnitude lower than one and no risk was therefore predicted for any of the investigated ENMs at the estimated release level in 2020. Compared to static models, the dynamic material flow model allowed us to use PEC values based on a more complex parameterization, considering a dynamic input over time and time-dependent release of ENMs. The PSSD approach makes it possible to include all available data to estimate hazards of ENMs by considering the whole range of variability between studies and material types. The risk assessment approach is therefore able to handle the uncertainty and variability associated with the collected data. The results of the current study are able to provide a scientific foundation for risk-based regulatory decisions of the investigated ENMs.

Natural toxins and harmful algal blooms (HABs): water and food safety, analysis, toxicity, and risks (I)

592 Occurrence of cyanotoxins in Greek lakes
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Cyanotoxins (CTs) produced from cyanobacteria in surface water during harmful algal blooms can pose significant threat to human health and the environment. Their analytical determination in both biomass and water is a demanding task as CTs comprise a large variety of compounds with different structural and physicochemical properties, i.e. cyclic peptides (microcystins, MCs and nodalins, NODs) and alkaloids (cylindrospermopsin, CYN, anatoxin-a, ANA-a and saxitoxins, STXs). The most important issues that make the CTs’ analysis challenging are firstly the large number of variants of various classes, the limited availability of analytical standards and insufficient validation data. Moreover, different methods of analysis are usually required for each class of CTs in order to achieve acceptable analytical performance. Based to the above there is a need to develop efficient multi-class/variant method protocols for analysis of as many as possible CTs. Our laboratory has recently developed and validated SPE-LC-MS/MS determination of multi-class CTs. As an example, an analysis of 12 MCs (D-Asp3-MC-RR, MC-RR, MC-YR, MC-HyR, -Asp3-MC-LR, MC-LR, MC-HiR, MC-WR, MC-LA, MC-LY, MC-LW and MC-LF), NOD, CYN and ANA-a in one run can be achieved. In addition, validated multi-variant methods for the analysis of STXs based on HILIC-MS/MS have been developed. Those methods have been combined in workflow to analyze multi-class toxins efficiently. The aim of this study was to demonstrate the applicability and efficiency of a proposed workflow for multi-class/variant determination of CTs. Futhermore, to detect and identify a wide range of CTs in Greek lakes never studied before, using this validated tool, Results of a monitoring survey in Greeks lakes showed that the proposed LC/MS/MS based workflow provided unequivocal and definitive identification of multi-variant/class toxins, avoiding the drawbacks of bioassay techniques that have been used previously. Using the proposed workflow a wide range of MCs (D-Asp3-MC-RR, MC-RR, MC-YR, MC-HyR, -Asp3-MC-LR, MC-LR, MC-HiR, MC-WR, MC-LA, MC-LY, MC-LW and MC-LF), NOD, CYN, ANA-a, STX and neoSTX were identified in Greek lakes for the first time. Acknowledgement: The authors thank CYANOCOST – Cost Action ES 1105 www.cyanocost.net

593 Interactions between cyanobacteria and daphnia
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Thanks to their adaptation cyanobacteria organized aquatic, marine and terrestrial environments. Eutrophication of waters has promoted and will increase cyanobacteria blooms in future, posing hazards to the aquatic ecosystem and human health due to the capability of cyanobacteria to produce bioactive or toxic compounds. One of the groups firstly affected by cyanobacteria is planktivorous zooplankton, such as Daphnia. On the other hand, Daphnia can also suppress cyanobacterial population up to certain density and toxicity. A development of tolerance apparently enables them to withstand cyanobacterial compounds, and is transferable to the next generation. The role of cyanobacterial toxins and other bioactive compounds has not yet fully been elucidated, neither has the question, if the presence of zooplankton grazers could modify their production. This study investigates the mutual two-way interactions, in terms of biochemical and life trait responses of both, cyanobacteria and daphnia. Microcystis aeruginosa PACC7806 culture was grown in a culture chamber designed, where two chambers are physically separated by a 0.2 μm cellulose nitrate membrane filter, preventing the grazing effect but allowing exchange of chemical compounds released into the medium. Exposures lasted one week. First results confirmed the detrimental impact of cyanobacterial metabolites released into their culture medium on D. magna. Cyanobacterial culture medium of M. aeruginosa PACC7806 obtained after 2 weeks culture, equivalent to 10^5 cells/mL, reduced feeding and survival, moreover altered detoxification and antioxidant response as well as the energetic budget. Exposures to spent media from M. aeruginosa PACC7806 micr- are currently in progress. Vice versa, Microcystis aeruginosa PACC7806 culture medium on D. magna cultures of two weeks: During the first days there was an increase of growth rates, followed by a decrease in physiological performance. Moreover, the antioxidant response increased, which, even though not significant itself, caused a significant reduction in the hydrogen peroxide content in the cyanobacteria. First results indicate that cyanobacteria not only harm aquatic organisms, but that vice versa they react to the presence of potential grazers, hence yet unknown substances present in the spent media impair their performance.

594 Teratogenic retinoid-like compounds produced by cyanobacteria into surface waters
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Acknowledgement

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developmental effects in zebrafish (Danio rerio) and frog (Xenopus laevis) embryos. Both the phenotypes and effective concentrations of exudates corresponded to all-trans retinoic acid (ATRA) equivalents, supporting the hypothesis that the teratogenic effects of cyanobacterial exudates are likely to be associated with retinoid-like activity. Non-target analyses and comparison of the spectra of compounds present in exudates with different retinoid-like activities pointed to structural features of compounds contributing to the retinoid-like activity. In the approach of a “virtual EDA” we have characterized in detail retinoid-like activity of 53 samples of exudates from 4 algae species and 15 cyanobacteria species/strains. Only several species of cyanobacteria showed detectable activity, while there was no activity in any algal exudates. We have identified a set of compounds that contribute to the total retinoid-like activity in both laboratory and field samples, including ATRA, 9/13cis retinoic acid, but also several novel cyanobacterial metabolites, such as 5,6epoxy-RA or 4keto-ATRA with high retinoid-like potency. In general, retinoid-like activity has been shown to be associated with cyanobacterial water blooms dominated by many different species, which documents that production of retinoids by cyanobacteria in the environment is a common phenomenon. This study was supported by the Czech Science Foundation Project No.18-15199S and FP7 SOLUTIONS project No. 603437.

595 (Co-)Production Dynamics of Cyanobacterial Peptides
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Cyanobacterial algal blooms are expanding temporally and spatially, which is promoted by eutrophication and likely climate change. Cyanobacteria can produce a wide range of bioactive compounds with different modes of action, including a variety of toxic algal peptides. Information on the formation of these novel cyanobacterial toxins is scarce. Here, we report the first research production dynamics of the majority of novel cyanopetptides is mostly unknown even for common cyanobacterial strains. Such information is crucial to assess the risk of these emerging natural toxins for human health in evaluating their potential to reach drinking water supplies. While it is known that many cyanopetptides are produced simultaneously from one species, the co-production of these potential toxins has not been explored comprehensively for cyanopetptides other than microcystins. This project focused specifically on the production and co-production dynamics of cyanopetptides from common cyanobacterial species including Microcystis aeruginosa and Anabaena flos-aquae strains. Batch culturing was done under varied environmental conditions and the cell abundance was followed by optical density, cell counting, and biomass was determined simultaneously; the cyanopetptides production was followed. Therefore, biomass extracts were purified and measured by liquid chromatography, high resolution mass spectrometry with refined analytical protocols. Comprehensive data analysis was performed to identify cyanopetptides and follow their abundance. These new insights of co-production dynamics are critical to better understand which peptides and peptide mixtures are present during cyanobacterial bloom events.

There are limited methods for the analyses of multiple algal toxins in aquatic food webs, phytoplankton, zooplankton, periphyton, macroinvertebrates, forage fish, bottom feeders and top carnivore fish. Algal toxins in freshwater systems do not necessarily occur as single contaminants; mixtures of toxins may be produced from Cyanobacteria, Prymnesium parvum (Prymnesins), and Euglena sanguinea, including microcystins, saxitoxins, cylindrospermopsin, anatoxin-a, prymnesins and euglenophycin. The objective of the first phase of this research was to spike extracts from plankton and fish homogenates with 3-5 dosages of microcystins (LR, LA and RR) individually and as mixtures, and to develop a method for their recovery and measurement using the MMPB derivatization method. The second phase of the project is to field-test this method on fish collected from water bodies experiencing algal blooms and compare results with individual congener measurements. Extraction methods and analytical methods being developed for this research are critical for development of procedures for plankton, periphyton, and macroinvertebrates. Ten and 100 mg of fish homogenates from fish containing 1, 4 and 14% lipids were spiked with 4 and 40 ng of each of the microcystin congeners, LR, LA and RR. Various extraction techniques and conditions were tested to optimize recovery and simplify the procedure. Overall toxin recoveries were found to range from 30 to 50%. The lipid content was found to not interfere with generation of MMPB; however, it did impact the workup/extraction procedure in ways which were accountable through the use of a surrogate standard. The MMPB technique can be reliably employed for microcystin recoveries were found to range from 30 to 50%. The lipid content was found to not interfere with generation of MMPB; however, it did impact the workup/extraction procedure in ways which were accountable through the use of a surrogate standard. The MMPB technique can be reliably employed for microcystin recovery and measurement using the MMPB derivatization method. The second phase of our study was to field-test this method on fish collected from water bodies experiencing algal blooms and compare results with individual congener measurements. Extraction methods and analytical methods being developed for this research are critical for development of procedures for plankton, periphyton, and macroinvertebrates. Ten and 100 mg of fish homogenates from fish containing 1, 4 and 14% lipids were spiked with 4 and 40 ng of each of the microcystin congeners, LR, LA and RR. Various extraction techniques and conditions were tested to optimize recovery and simplify the procedure. Overall toxin recoveries were found to range from 30 to 50%. The lipid content was found to not interfere with generation of MMPB; however, it did impact the workup/extraction procedure in ways which were accountable through the use of a surrogate standard. The MMPB technique can be reliably employed for microcystin quantification in fish tissue. Detections in non-spiked samples (10-20 ug/kg) are comparable to literature precedent. For tissue quantification the MMPB method provides considerable improvements over extraction of individual toxin congeners and is consistent even with very polar or hydrophobic MCs.

597 Saponins in the aquatic environment: hydrolysis and toxicity
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Saponins are a class of bioactive natural compounds. Due to their detergent-like structure, saponins have a lot of applications, e.g. as biopesticides in crop protection. They may leach into the aquatic environment due to their low octanol/water partition coefficient and poor binding to organic matter. They may therefore pose a risk to the aquatic organisms. However, their fate and toxicity in the environment are not fully understood. Hence, we aim to investigate the pH and temperature dependence of saponins hydrolysis together with their dissipation in sampled lake waters and to determine the aquatic toxicity of saponins from quillajka bark, tea seed coat, and quinoa seed coat towards different aquatic organisms. The hydrolysis of saponin (quillaja saponin) showed to be a highly pH dependent base-catalyzed reaction. The half-life was around 330 ±220 days at pH 5.1 and 26°C, while decreased to 0.06 ±0.01 at pH 10.0. The hydrolysis was also influenced by temperature with an activation energy of 56.9 ±14.2 kJ/mol at pH 7.2. Lake waters with pH varying between 6.4 and 8.2 showed completely different hydrolysis patterns, with a fast initial dissipation of up to 60% of the initial saponin concentration, followed by an extremely slow to nil reaction, making saponin partially persistent in lake waters. The maximal concentrations protecting 95% of the aquatic species (HC95) derived from the SSD’s of saponins from quillajka bark, tea seed coat, and quinoa seed coat were 2.91 ±0.00, 0.22 ±0.11 and 22.9 ±5.84 mg/L, respectively. The 100-fold difference in toxicity between the saponin-rich extracts from different plant species indicate that saponin toxicity depends on the species where it origins from, making “read-across” between saponins a dubious exercise. In addition, the predicted environmental concentrations of different saponins are close to or higher than their water quality standard, which means that the saponins might pose a risk to the aquatic environment if not used cautiously. Therefore, we recommend not using surrogate or expected data/conclusion in the regulation of saponin-rich plant extracts and pay more attention to the potential risk of saponins to the aquatic environment.

Advancing the Adverse Outcome Pathway Framework - An International Horizon Scanning Approach
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Recognizing the international interest surrounding the adverse outcome pathway framework, which captures existing information describing causal linkages between a molecular initiating event through levels of biological organization to an adverse outcome of regulatory significance, an effort was undertaken to provide the scientific community the opportunity to engage in determining the direction of the AOP framework. Specifically, a horizon scanning effort was used to solicit questions from the international scientific community asking participants to propose questions that consider key outstanding challenges and/or limitations that must be overcome to advance the AOP framework for both research and regulatory decision making. From March-June, 2016, 340 valid questions were collected from 158 global submissions, spanning all continents, to an online horizon scanning survey. Respondents to the survey self-identified as 35% academia, 35% government, 20% industries and other stakeholders and 10% from the public. After receiving question solicitation, questions were separated into broad topic areas including, AOP networks, quantitative AOPs, collaboration and communication on AOPs, AOP discovery and development, extrapolation, exposure/toxicokinetics considerations, and AOP application. An expert-ranking exercise was then conducted to identify high-priority questions for each category and from this, four broad themes emerged including future AOP research and regulatory initiatives. These themes were used as workgroup topics for a Pelliston™ Workshop, including: AOP networks and their applications; quantitative AOPs and
her their applications; regulatory use of the AOP framework, and expanding awareness of, involvement in; and acceptance of AOPs to support aspects of predictive toxicology and regulatory decision-making. Charge questions for each workgroup were directly modified from those submitted during horizon scanning. Additionally, from the horizon scanning exercise, frequently asked questions (FAQs) were identified and addressed by experts in the field. Together the horizon scanning, expert conducted exercise, and questions to FAQs are used to set the stage for the SETAC Pellston Workshop titled “Advancing the Adverse Outcome Pathway Concept: An International Horizon Scanning Approach,” that took place in Cornwall, Canada during April 2017. The contents of this presentation neither constitute nor necessarily reflect US EPA policy.

599 Adverse Outcome Pathway networks: development, analytics and applications D. Knapen, University of Antwerp / Zebrafish Dept Veterinary Sciences; M. Angrish, US EPA; National Center for Environmental Assessment; M.C. Fortin, Alcami / Environmental and Occupational Health Sciences Institute; I. Katsiaziadaki, Cefas / Environmental and Animal Health; M. Leonard, BORéal SA, L. Margiotta-Casaluci, Brunel University London / Institute of Environment, Health and Societies; S. Munn, European Commission; J. O'Brien, Environment and Climate Change Canada / National Wildlife Research Centre; N. Pollesch, US EPA / ORD NHEERL Mid Continent Ecology Division; L. Smith, University of Florida / Physiological Sciences; X. Zhang, Nanjing University / Environmental Science; D. Conolly, US EPA / National Health and Environmental Effects Research Laboratory

Adverse outcome pathways (AOPs) are an important framework that can help support more effective use of mechanistic, pathway-based, data in risk assessment and regulatory decision-making. AOPs have rapidly evolved from a conceptual paradigm into a formalized framework for organizing biological and toxicological knowledge. In the current context, it is generally accepted that in response to the recognized need to continue advancing the framework, SETAC sponsored a global horizon scanning exercise to identify major outstanding topics and challenges related to the AOP framework and its application. The development of guidance related to AOP network development and analysis was identified as a critical need. This report captures the expert consensus work, and answers to FAQs, also on related topics such as mixture toxicity assessment and the implementation of feedback loops within the AOP framework. This presentation briefly outlines critical concepts concerning the development of AOP networks, how they may be analyzed, and illustrates how information derived from them can be applied. First, derivation of AOP networks is considered in the context of how it differs from traditional dose-response modeling. Next, the application of filters and layers is discussed, which can be used to refine and enrich derived AOP networks so that they may be tailored to address specific questions of interest. We then introduce a number of analytical and computational approaches that may be used to characterize and analyze the structure of AOP networks to derive information that can guide research and regulatory decision-making. A number of application case studies is used to illustrate concepts underlying development and analysis of AOP networks, and how those concepts tie in with ultimate application. The contents of this presentation represent the personal opinions of the authors and neither constitute, nor necessarily reflect the policies or viewpoints of their employers or institutes.

600 Building and Applying Quantitative Adverse Outcome Pathway Models for Chemical Hazard and Risk Assessment S. Scholz, Helmholtz Centre for Environmental Research / Department Bioanalytical Ecotoxicology; R. Ashauer, University of York / Environment; R. Conolly, US EPA RTP; B. Landesmann, JRC, European Commission; I. Katsiaziadaki, Cefas / Environmental and Animal Health; M. Leonard, BORéal SA, L. Margiotta-Casaluci, Brunel University London / Institute of Environment, Health and Societies; S. Munn, European Commission; J. O’Brien, Environment and Climate Change Canada / National Wildlife Research Centre; N. Pollesch, US EPA / ORD NHEERL Mid Continent Ecology Division; J. Wheeler, Dow AgroSciences; A. Zapanic, Eawag Swiss Federal Institute of Aquatic Science and Technology / Environmental Toxicology; E.J. Perkins, US Army Engineer Research and Development Center / Environmental Laboratory Quality Assurance; J. Eichhorn, Department of Environmental Health and Environmental health is important in understanding the potential hazards and risks of using, or being exposed to, chemicals. Here we examine how the Adverse Outcome Pathway (AOP) concept and knowledge base can be used to develop quantitative models (qAOPs) to predict and assess hazards and risks of chemicals. Quantitative models can be developed with a clear problem definition and using AOPs as initial data. Models and methods range from semi-quantitative to quantitative modeling approaches or combination of these (e.g., fully mechanistic mathematical / ordinary differential equation based, individual-based models, statistical, or Bayesian network models). We discuss best practices for choosing modeling approaches, model building and the necessity for transparent and comprehensive documentation in order to gain confidence in the use of a model. Finally, we present examples of how qAOP models can support decision making: a screening level assessment of the health hazards of chemicals and chemical mixtures using a qAOP Bayesian network model of stoichiostasis, the use of qAOPs in a prospective risk assessment context (e.g. in vitro to in vivo extrapolation using aromatase inhibition as an example) and for extrapolation between species or life stages.

601 Use of Adverse Outcome Pathways to Inform Decisions on Chemical Innovation, Regulation & Stewardship S. Hill, US EPA NHEERL / ORD NHEERL Integrated System Toxicology Division; P. Browne, OECD / OSCP; K.K. Coady, The Dow Chemical Company / Toxicology Environmental Research and Consulting; M.R. Embry, ILSI Health and Environmental Sciences Institute (HESI); T. Hutchinson, School of Biological Sciences, Plymouth University / School of Biological Sciences; E. Lienala, OECD; L. Maslankiewicz, National Institute for Public Health and the Environment (RIVM); T.M. Steeger, U.S. EPA / Office of Chemical Safety and Pollution Prevention

An invited group of scientists participated in a SETAC Pellston Workshop titled “Advancing the Adverse Outcome Pathway Concept – An International Horizon Scanning Approach,” in April 2017. The workshop addressed key challenges or limitations of AOP constructs as tools for informing research and regulatory decisions that were identified by responses to a global Horizon Scanning survey. This presentation will summarize the findings of Pellston Workgroup 3, which was tasked with the explicit justification of specific considerations for this use of AOP constructs in regulatory decision making. The use of AOPs and related concepts have increased in scientific and regulatory sectors over the past decade, coinciding with pressures to find innovative solutions to evaluate chemical safety in a more efficient and effective manner that better directs resource utilization. This workshop focused on how AOPs can be a useful tool for chemical decision makers in the government and private sector. At the various points where chemical decision making is employed across the “life” of a chemical – from research and development within the commercial sector, government registration and regulation, through to post-marketing use/stewardship – AOPs can be used as an organizing principle. Pragmatic evidence is provided for how AOPs can be and are currently being used in chemical decision-making processes. Considerations for evaluating the suitability of AOP for decision makers are discussed, recognizing that the acceptable level of uncertainty varies based upon the nature of the decision and the context in which it is being applied. The presentation provides multiple examples of AOP use and practical considerations for evaluating whether use of AOPs is fit-for-purpose in different circumstances. This abstract does not necessarily represent the views or policies of the U.S. EPA.

602 Ensuring Long-Term Utility of the AOP Framework and Knowledge for Stakeholders G.T. Ankley, U.S. EPA / National Health and Environmental Effects Research Laboratory; A. Carusi, University of Sheffield; H. Davies, WA State Dept of Health / Dept of Ecology; G. De Grandis, Norwegian University of Science and Technology; B. Escher, Helmholtz Centre for Environmental Research GmbH - UFZ / Cell Toxicology; G. Hodges, Unilever Research / Safety and Environmental Assessment Centre; K. Leuwer, The University of Hong Kong / The Swire Institute of Marine Science and School of Biological Sciences; M. Whelan, University of Leicester / Geography; C. Willett, the Humane Society of the United States / Animal Research Issues

The adverse outcome pathway (AOP) framework serves as a knowledge assembly and communication tool to facilitate translation of mechanistic (e.g., molecular, biological, histological) adverse apical outcomes meaningful to chemical risk assessment. Although initially designed for ecotoxicology applications, the framework has also received extensive attention relative to chemical safety assessments for human health. Moreover, as the AOP concept and associated knowledgebases have evolved, it has become recognized that the potential stakeholder community is broader than scientists and regulators directly involved in chemical safety assessment. For example, the application of AOP-based thinking for addressing biomedical challenges has become increasingly evident. This presentation will identify various stakeholders who currently, or could potentially, benefit from application of the AOP framework and knowledge to specific needs, and describes challenges and strategies to effectively engaging these stakeholders. We also present a “roadmap” on how to maintain a viable, sustainable network to support AOP stakeholders, including recommendations for governance and coordination of AOP development and knowledge dissemination in a multi-stakeholder consortium. The contents of this abstract neither constitute, nor necessarily reflect, official USEPA policy.

603 Adverse Outcome Pathways: Moving from a scientific concept to a globally accepted framework M. Hecker, University of Saskatchewan / School of the Environment & Sustainability and Toxicology Centre; C. LaLone, U.S. EPA / Mid Continent Ecology Division

The adverse outcome pathway (AOP) framework has gained significant international traction as a systematic approach for capturing existing knowledge to transparently link mechanistic data to apical toxicity endpoints as a means to inform research and risk assessment. While the framework has evolved significantly since
its introduction in 2010, it was recognized that a survey of the broader scientific community would be useful in identifying shortcomings and in guiding future initiatives. In 2016, we reached out to national and international scientific and regulatory communities to collect questions and provide an opportunity to discuss key outstanding challenges that must be addressed in order to realize the full potential of the AOP framework. Four key themes emerged from this “Horizon Environment” exercise (see presentation “Advancing the Adverse Outcome Pathway Framework – An International Horizon Scanning Approach” in this session), which were then addressed at a Society of Environmental Toxicology & Chemistry (SETAC) PellanSTM Workshop comprised of international participants representing industry, government, academia, and NGOs was held in Cornwall, Ontario, in April 2017. This presentation will provide an overview of the overall outcomes and common themes that emerged during this Pellan Workshop. In brief, common themes that spanned across these main topics included the need to simplify, translate, and better communicate the AOP framework to the broader international stakeholder community, and a consensus that the AOP framework does not represent a rigid tool but rather a knowledge repository for diverse stakeholders ranging from epidemiologists to mainstream experimental toxicologists to regulators and managers. Furthermore, while considering the framework and its applications, the field of environmental toxicology and human health naturally merged into a continuum that is at the nexus of Toxicology in the 21st century. In particular, it was felt that the current momentum the AOP framework has gained across a wide range of professional sectors provides the unique window of opportunity to reach out to and gain acceptance of this framework by society, which will be required for it to become an integral part of the international chemical and environmental risk assessment landscape. The contents of this presentation neither constitute nor necessarily reflect US EPA policy.

Environmental specimen banks in research and regulation for a better environmental quality

604 Monitoring of POPs in the Swedish aquatic ecosystem and in human milk E. Nyberg, A. Bignert, S. Danielsson, Swedish Museum of Natural History; C. Ek, Department of Applied Environmental Science / Department of applied environmental science.

In the 1960s, the Baltic Sea was found to be severely polluted by persistent organic pollutants (POPs). These discoveries were the starting point of a continuous Swedish national monitoring program for contaminants in biological matrices, mainly from the marine and freshwater environment, with samples dated as far back in time as 1969. Today’s marine and freshwater monitoring programs consist of 32 lakes and 28 marine sites where matrices as perch, pike, arctic char, herrings, cod, eelpout, blue mussel and eggs from guillemots, oystercatcher and common tern are annually collected. In 1967, examination of human exposure to POPs was initiated by Karolinska Institutet in Stockholm through measurements in human milk from the area, and since 2007 milk has also been collected from Gothenburg in the southwest of Sweden. The milk samples were in 1997 transferred to the Environmental Science Institute at the Swedish Museum of Natural History.

The main objectives of the monitoring program are to investigate changes over time, to examine geographical differences and to assess compliances with set target values. Moreover, the program is designed to answer these different questions with a high statistical power. Since the start of the monitoring, concentrations of PCBs, DDTs, HCHs and HCB have decreased in fish and bird eggs from both the Baltic Sea and the Swedish freshwater environment. Several of the classical POPs have also decreased considerably in human milk. However, the non-linear trends differ between the monitoring matrices for several contaminants. In some cases the peak differs, and in others, concentration is levelling out for one matrix but continues to decrease for another. In addition, the concentrations of PCBs, DDTs and HCHs are, despite continuous decreases since the 1970’s, still higher in the Baltic Sea compared to, for example, the North Sea.

605 Jumping out of the frying pan and into the fire? Spatial and temporal trends for PBDE, Dechlorane Plus and alternative flame retardants in samples of the German Environmental specimen bank A. Dreyeg, Eurofins GFA GmbH / Air Monitoring; F. Neugebauer, Eurofins GFA Lab Service GmbH / R&D; N. Lohmann, Eurofins GFA LabService GmbH; M. Paulus, Trier University; H. Ruedel, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Department of Environmental Monitoring; C. Rauet, Umweltbundesamt / International Chemicals Management; J. Korschoreck, Umweltbundesamt.

In the last century, conventional brominated flame retardants (FRs) such as polybrominated diphenyl ethers (PBDEs) were identified as persistent organic pollutants and subsequently regulated. Novel or alternative FRs were introduced as their replacements to meet ongoing market demands. Many of these alternative FRs are also highly chlorinated or brominated and their fate and effects in the environment may be similar to those of their regulated counterparts. Until now there are only few comprehensive data sets about alternative FRs in the environment, particularly for Germany. In order to provide for a systematic overview about the current state of contamination of the German environment to FRs, a large set of terrestrial, freshwater and marine samples from the German environmental specimen bank were analysed for 45 FRs (PBDEs, Dechlorane Plus and brominated aromatics, brominated ethers, cyclic BFRs). The substances will be discussed with respect to their spatial occurrence in the environment (including different matrices as well as land use and ecosystem types), their substance patterns in food webs and the temporal evolution going back to the 1980s, e.g. from coastal herring gull eggs, freshwater fish, tree leaves and roe deer will be used to illustrate time trends for regulated flame retardants and their substitutes. Recommendations will be given to European and international chemical management.


The National Institute of Standards and Technology (NIST) has been involved in the long-term archival of biological and environmental specimens for over 40 years. Specimens originally intended for monitoring geographic and temporal trends in emerging contaminants as well as changes in transport and accumulation of legacy contaminants have added value today. Tissue and fluid specimens from marine animals, including mammals, seabirds, sea turtles, bivalves, fish, coral and coral ecosystems, collected through various projects are archived at the Marine Ecosystems Specimen Bank (MBS) in the NIST Harry Diamond Laboratory, in Charleston, South Carolina, USA, using standardized protocols for collecting, processing, and cryogenic storage. The protocols ensure a high quality sample is provided for downstream analysis that is fit-for-purpose and that homogenous aliquots are uniform, reproducible, and stable over time. New investigations exploring if the standardized protocols, 1) affect the quality and suitability of RNA for downstream expression studies conducted in our laboratory, 2) facilitate feasibility of a forensics-style approach for taking indoor air and dust samples from two European ESBS. Air samples were taken using passive air samplers while dust samples were taken using wet wipes on horizontal surfaces (not floors). Four room types were included at each ESB; main laboratory, changing room, storage room, and a corridor. The results show that several of the non-regulated CECs are found at high levels in the indoor matrices. These include: chlorinated paraffins, new flame retardants, and siloxanes. In contrast, regulated contaminants (e.g. polychlorinated biphenyls, PCBs, polybrominated diphenyl ethers, PBDEs) are found at low levels. Weather the high levels observed in indoor air and dust affect the ESB samples or not is to be further studied but the findings highlight an important aspect to take into consideration in the QA protocols of ESB.

607 Monitoring of the indoor environment of ESB laboratories with selected target and non-target screening methods P. Bohlin Nizzetto, Norwegian Institute for Air Research; M. Schlabach, A. Halse, P. Rostkowski, NILU Norwegian Institute for Air Research

The environmental specimen banks (ESBS) handles and store a wide range of high-quality samples with a main objective to perform retrospective analyses of newly identified contaminants (chemicals of emerging concern, CECs). It is of highest importance that the ESBS can be used to illustrate the current state of contamination of the environment and not contaminated during handling and storage. Strict quality assurance (QA) protocols are applied at the individual ESBS, including specially dedicated laboratories, no use of personal-care products, cleaning routines and more. More complicated to control is the indoor air and dust in the laboratories and storage locations. Building materials and indoor objects may contain and further enhance the concetration of CECs to the indoor environment. To evaluate trends in concentrations of perfluorinated alkyl acids (PFAAs) retrospectively, using samples stored and processed in polytetrafluoroethylene (PTFE) based materials, have recently been conducted. In addition, alternative uses of these cryopreserved specimens including, 1) a detailed proteome profiling of tissues used to evaluate a new high-coverge well- scaffolded genome, and 2) the discovery of using total DNA as an alternate modern tool to genetic species identification, have also been conducted and will be discussed.

608 DNA banking and its relevance for biodiversity research J. Astrin, Zoological Research Museum Alexander Koeing

Within their genomes, the organisms on our planet contain an immense wealth of information about the diversity of life. These genomes conserve the code to identify organisms, comprehend population structures, etc. Fast progress in molecular technologies dramatically speeds up research on genetic biodiversity and increases the demand for professionally preserved and managed genome-quality samples in many disciplines, e.g. in ecology, conservation biology, etc. Biodiversity biobanks cater specifically to these demands, and in a standardized way. Environmental samples collected periodically by ESBS following defined routines constitute a very valuable source of DNA for biodiversity research, as they keep open a window that allows the parallel, correlative analysis of the chemical and of the species community composition of a given environment over time. Through species
identification via DNA barcoding and high throughput sequencing, such samples can be most helpful in documenting and interpreting environmental change. Accumulated snippets of free DNA in soil or water samples (freshly collected or from ESBs), so-called environmental or eDNA, enable the comprehensive appraisal of species compositions in a multitude of environments. DNA extracted from ESB samples should be deposited in dedicated DNA banks in order to make this resource available to the scientific community and the evolving DNA analysis methodologies (which may prompt repeated analyses of samples over time, with ever-increasing amounts of genomic data recovered). Thus, by adding DNA banking to their service spectrum, ESBs can considerably increase their visibility and public demand. Furthermore, they can foster knowledge aggregation at the biodiversity level around their ESB samples, making these more valuable.

609 Discussion on environmental specimen banking in research and regulation

Hazard and exposure assessment of chemical mixtures: steps towards increasing the realism of chemical risk assessment (II)

610 Poster spotlight: TH273, TH288, TH285

611 Environmental risk assessment of multiple stressors - chemicals and ionizing radiation

K. Petersen, NIVA - Norwegian Institute for Water Research; J. Brown, Norwegian Radiation Protection Authority; K. Tollefsen, NIVA / Ecotoxicology and Risk Assessment

Increased focus on cumulative effects of pollutants in the environment has led to development of several methods for environmental risk assessment (ERA) of chemical mixtures and for ionizing radiation. Even though no generic impact and risk assessment model exists to accommodate different types of stressors (e.g. multiple stressors such as radiation, metals and organic compounds), larger harmonization and integration of approaches taken can be achieved to improve and combine the existing models for ERA of chemical mixtures and for ionizing radiation from radionuclides. Here we present a potential 2-tired approach for risk assessment of multiple stressors by assuming additivity of chemicals and ionizing radiation as a first approach in order to combine a framework for ERA of chemical mixtures with the ERICA tool for ERA of radionuclides. The proposed approach was applied to a real case scenario: emission from decommissioning of old oil platforms performed on-shore close to Vatsfjorden (Norway). Several metals, NORMs and organic pollutants are monitored as part of the activity. Effect data for the monitored compounds were compiled from various databases and literature. The Tier 1 identified a cumulative environmental risk of the stressors, and several metals and organics had a risk quotient above 1 (preliminary data). The potential for a cumulative environmental risk was verified in Tier 2 where species group specific risk was investigated. Metals were identified as the main risk drivers for algae, crustaceans and fish, where fish was identified as the most sensitive species group for this exposure scenario. Based on the used exposure scenario, compiled effect data and the suggested approach for ERA of multiple stressors, a potential environmental risk was observed. The main challenges and uncertainties for the proposed approach are linked to exposure data in terms of speciation and bioavailability; time consuming and subjective effect data compilation and assumption of additivity of ionizing radiation and chemical stressors. The approach for estimating environmental risk of multiple stressors requires validation through experimental studies, but could already serve as a suitable tool for prioritization of stressors and organisms of concern, and to identify knowledge-gaps in terms of exposure and effect data. Acknowledgements: The project was funded from NRC project 223268 (CERAD) and in-kind from Norwegian Institute for Water Research (NIVA)

612 Assessing health risk associated with micro-pollutant mixtures in drinking water: an innovative combination of in vivo and in vitro assays and analytical screenings

Y. Levi, Univ. Paris Sud / ESE UMR 8079; J. Loret, o. schlosser, CIRSEE Suez Environment; a. guillon, Suez / CIRSEE; m. favier, INSERM Institut Cochin; v. domergue davenport, Université Paris Sud; M. Binbot, V. Huteau, Univ. Paris-Sud / UMR ESE; m. plewa, b. marinas, University of Illinois at Urbana Champaign Assessing health risks associated with organic micropollutants in drinking water is a major challenge for public health and improvement of the drinking water production units. Given the low concentrations, the diversity of emerging contaminants and chronic exposure, it is essential to combine chemical analysis with biotests. In vitro bioassays or in vivo tests on aquatic animals allow effect screenings but interpretation for health risk assessment is difficult. We have designed an innovative research program (to our knowledge, the first of its kind applied to drinking water) that aimed to simulate human life-long exposure to micro-pollutants in drinking water for in vivo tests on mammals... Over 4 seasons, we have concentrated by 100 the organic fraction from large volumes of 4 types of water: river water, drinking water produced by a treatment plant with this raw water, reverse osmosis distillate water and mineral water. 2 generations of mice (10 males, 10 females for each type of water) were fed with contaminated drinking water during a 1 year period, in order to reproduce in the animals, the exposure as a man drinking the water over 70 years. We carried out complete chemical analysis (quantitative targeted analysis, innovative non-targeted screening by HPLC and GC-2D with mass spectrometry), offering a new vision of the contaminants diversity. In vitro bioassays were performed on mammalian cell lines (ER and AR recept.), cell toxicity tests (mammalian and Microtox) and genotoxicity on CHO cell assay. Mice growth, survival rate and behavior in open field and in elevated plus maze were studied. Histological analysis on 19 organs and blood hormonal assays were performed. This complete and innovative protocol did not show significant difference of survival rate and growth between the four mice groups. In contrast, histological and hormonal effects were observed in the mice exposed to the raw river water concentrates as opposed to the mice exposed to the waters of better quality. Several tests suggested a greater sensitivity of the 2nd generation mice as compared with the 1st generation. Biotests confirmed the overall good quality of the treated water, whatever the treatment process, despite the presence of organic micropollutants. The presentation will show the detailed protocol of the major microalgae, Phaeocystis antarctica and Cryothecomonas armoricana. The determination of their health risk assessment with regard to long-term exposure to real mixtures of organic pollutants in drinking water.

613 The application of DGT to assess the risk of metal mixtures in polar environments

D. Koppel, University of Wollongong / Chemistry; M.S. Adams, CSIRO; C.K. King, Australian Antarctic Division; D.F. Jolley, University of Wollongong / School of Chemistry

Contaminants predominantly occur in mixtures, posing a challenge to environmental management practices which are usually based on single-contaminant toxicity. Chemical interactions of the contaminants and non-specific biological responses to these mixtures may result in effects that differ from the sum of the toxicity of individual components. These differences can be classed as antagonism (less toxicity than expected from the sum of the individual contaminants in the mixture), non-interaction (toxicity equal to that expected from the sum of the individual contaminants), synergism (more toxicity than expected from the sum of individual contaminants). Diffusive Gradients in Thin-films (DGT) has been established as a robust method for analysing the biologically-available contaminants in situ and is well-positioned to assess the toxicity of contaminant mixtures. This study explores the use of DGT (with a Chelix-100 resin) to assess the toxicity of Cd, Cu, Ni, Pb, and Zn in mixtures, to two common Antarctic marine microalgae, Phaeocystis antarctica and Cryothecomonas armoricana. Non-interactive and synergistic toxicity were observed in the two algal species in response to increasing multiples of an environmental mixture (where the ratios of metals were based on reported concentrations at a historically contaminated Antarctic marine bay). Non-interactive toxicity was observed in response to an equitoxic mixture (five metals at their EC10 concentrations), as determined by Independent Action and Concentration Addition modelling. DGT measured concentrations were able to predict toxicity to the two algae at environmentally realistic contaminant concentrations. The use of DGT in Antarctica was validated by field deployments to sediments and waters of the near-shore coastal environment of a contaminated site around an Australian Antarctic research station. Their use in assessing the risk of historical contaminants to the polar environment is discussed.

614 Ecotoxicity testing of environmentally realistic contaminant mixtures using passive samplers: what can we learn from repeating toxicity tests over an extended period of time?

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In current environmental risk assessment, researchers assess effects caused by single substances to single species and extrapolate for realistic conditions where organisms are usually exposed to complex contaminant mixtures. The use of passive sampling opens new possibilities to work with such mixtures and to transfer them into biotest systems by either applying passive dosing (for equilibrium based samplers) or extract spiking (for integrative samplers). Our research objective was to investigate whether or not environmentally realistic contaminant mixtures
has effects on marine phytoplankton and how effects could be explained through the analysis of results, we propose some lead and improvements for the soil community risk assessment. The multivariate analysis (PRC) is used for the higher tier studies is the low number of replicates which could lead to false negative or false positive. The recently improved Minimum Detectable Difference is used to assess the robustness of the data used in these statistical tests. Then, through the analysis of results, we propose some lead and improvements for the soil community risk assessment, from the experimental design to the sampling choice and statistical analysis in the context of higher tier regulatory risk assessment of chemicals for earthworm and soil communities.

618 Metal soil threshold calculator tool: use of available data for derivation of metal soil quality standards for different scenarios and protection goals K. Oorts, INTERACT; E. Scholz, INTERACT; J. Chowdhury, International Lead Association / Senior Scientist - Environment During the last 2 decades, intensive research has been performed to improve the risk assessment of metals in soil and numerous chronic metal toxicity data were generated for various terrestrial species and microbial functions in different soil types. Models were developed for correction for differences in bioavailability among soils and for differences between laboratory and field conditions. For many metals, the large amount of chronic toxicity data for different species and functions allows the application of the species sensitivity distribution approach (SSD) to derive soil quality standards. This work was mainly triggered by the European legislation on chemical management (REACH) and the data were therefore primarily used to derive more realistic effect concentrations (PNEC) for respective risk assessment. To facilitate a more flexible derivation of ecological quality standards for metals in soil for different protection goals (e.g. remediation thresholds), jurisdictions, regions or sites, while still making maximal use of the wealth of data and models already available, a metal soil threshold calculator tool has been developed. This freely available spreadsheet reports almost 1200 reliable toxicity data for a wide range of metals (Cd, Co, Cu, Pb, Mo, Ni and Zn to soil organisms (plants, invertebrates and microbial processes)) and calculates ecotoxicological threshold concentrations expressed as (pseudo-)total (i.e. aqua-regia extractable) metal concentrations in soil (mg/kg dry weight). All metals covered have sufficient chronic toxicity data allowing the derivation of an SSD. The soils used for ecotoxicity testing cover for each metal a wide range of soil properties, making the results representative for most regions in the world. Several optimal options are included to allow calculation of metal soil threshold concentrations for various goals (e.g. risk assessment or setting of remediation thresholds for different land uses): selection of organism groups or species to be considered, selection of 

Advances in Soil Ecotoxicology and Risk Assessment of Terrestrial Ecosystems (II)

616 How protective is the current risk assessment for soil invertebrates? P. Kotschik, Umweltbundesamt / Federal Agency of Environment / Risk assessment for plant protection products; J. Roembke, ECT Oekotoxikologie GmbH; J. Sousa, University of Coimbra / Department of Life Sciences; T. Natal da Luz, National Laboratory of Geophysical Sciences, University of Coimbra; S. Chelinho, CFE Centre for Functional Ecology / Department of Life Sciences of University of Coimbra; N. Capela, CFE Centre for Functional Ecology; S. Pieper, German Federal Environment Agency (UBA) / Plant Protection Products The present risk assessment evaluating effects from plant protection products (PPP) as well as other chemicals towards soil organisms encloses uncertainties. According to the recently published report on terrestrial ecotoxicology, the risk assessment for soil organisms exposed to PPP is based, in a first step, on results of standardised tests performed in artificial soil. This substrate is a mix of sand, peat, kaolinite clay and calcium carbonate and is not comparable to natural soils. However, limited data is available on the degree to which soil parameters influence the toxicity of chemicals for soil invertebrates in field soils, since toxicity is modulated by chemical sorption and bioavailability. Possibly, soil parameters like pH, organic matter content or texture, interact with each other in affecting the toxicity of different compounds to different soil organism groups. These uncertainties in the accurate estimation of the extrinsic lab to field effects might be addressed by the investigation of the toxicity of test chemicals for organisms in natural soils. A screening project has been initiated in spring 2016, comprising a literature review aiming to investigate the effects of soil properties on the impact of PPP’s on soil organisms. The practical part of the project included laboratory studies on Eisenia sp. and Folsomia sp. on 4 PPP in 5 different soils. The results showed deviations on toxicity values obtained for single substances in different soils up to factor 6. The highest differences were detected for Folsomia sp. exposed to the active substance Pendimethalin in OECD10% and Lafa 2,1, respectively. So, the current risk assessment schemes for soil organisms based on standard laboratory studies performed with the surrogate species Eisenia sp. as well as Folsomia sp. is not always protective. Uncertainties in the assessment do exist regarding the effects of chemicals in natural soils towards other soil organisms, and other varying soil parameters that have not been investigated systematically so far (e.g. pH, clay content and interaction between them). The conducted literature research as well as the performed laboratory studies should be classified as preparatory work for more comprehensive studies. Focus of the future research should be set on the identification of key parameters influencing toxicity in different groups of species in higher tier conceptual models allowing the extrapolation from the lab towards the field situation.

615 Marine Diatom Exposure to a Complex Mixtures of Fourteen Chemical Pollutants at Environmental Concentrations. What did we learn? D. Napierska, European Commission Joint Research Centre / Directorate D Sustainable Resources Water and Marine Resources Unit; R. Carvalho, European Commission Joint Research Centre / Directorate D Sustainable Resources Water and Marine Resources Unit; I Sanseverino, European Commission Joint Research Centre; S Balzamo, M. Potalivo, ISPRA Institute for Environmental Protection and Research; R. Loos, European Commission Joint Research Centre; D. Marinov, T. Letteri, European Commission - Joint Research Centre / Directorate D Sustainable Resources Water and Marine Resources Unit Thousands different chemicals are discharged into the environment from agriculture, industry, medical facilities, house-holds. Currently, there is an increasing concern for the environmental impact of mixture of compounds since the additive and eventual synergistic effects are unknown and could produce serious adverse effects. To address this issue, a joint effort of 16 European and associated research groups participated to an exercise to test a 14-substance synthetic reference mixture at safety environmental concentration under the Water Framework Directive (Environmental Quality Standard, EQS). The mixture, was tested on the own routine bioassays to investigate the chemical mixtures effects (Carvalho et al., 2014). The bioassays covered the entire ecosystem from bacteria to fish as well in vitro assays providing an unique scenario from ecological risk assessment perspective. The results showed that effects were observed at very low concentration on algal-bacteria composition in a marine microcosm, immobilization in crustacean, fish embryo toxicity and frog embryo development. Transcriptomics analysis was performed for the marine diatom Thalassiosira pseudonana exposed either to single compound or the mixture to investigate whether the single exposure and multiple exposure would show different gene expression profile pattern. The results show that the mixture induces a pattern similar to the ones induced by the single herbicides Diuron and Isoproturon. Signatures induced by the Cadmium or Nickel partially overlapped with the mixture signature. The exposure to the other compounds did in general not induce relevant signatures, although a weak overall consistent signature is present for some of them. In conclusion the effects of the mixtures could be explained mainly by the exposure to the two herbicides.
effect levels from the original dose-response curves (EC$_{x}$), selection of protection level (probability level in SSD), bioavailability models to be included etc. The advantages of this approach are the maximal use of available toxicity data and bioavailability corrections and the enhanced transparency in the derivation of ecological quality standards for metals in soil for different goals and different scenarios.

619 Assessment of pesticides on a landscape level - What is basically needed? A. Toschki, Research Institute gaiac; M. Hammer-Wirtz, Research Institute gaiac / gaiac - Research Institute for Ecosystem Analysis and Assessment; B. Scholz-Starke, M. Ross-Nickoll, RWTH Aachen University, Institute for Environmental Research / Institute for Environmental Research

It is recently discussed and is widely accepted that species diversity and habitat quality dramatically decreased in the last decades due to the increasing intensity of agricultural land use. The decline was proved for various species groups, such as soil arthropods, insects, butterflies, birds etc. With the loss of species also a variety of benefits and ecosystem services that were provided by the species dropped away. The intensive use of pesticides has not only affected the biodiversity in the field and possible effects of the combined or sequential use. Additionally, there is no transparent documentation of the loss of biodiversity in the field and possible effects of the combined or sequential use. The intensive use of pesticides has not only affected the biodiversity in the field and possible effects of the combined or sequential use. The intensive use of pesticides has not only affected the biodiversity in the field and possible effects of the combined or sequential use.

Wood-fibres composite in substitution of a synthetic material to enhance sustainability purposes for automotive sector S. Maltese, Magneti Marelli SpA / Powertrain Division; a. bonoli, DICAM- Alma Mater Studiorum - University of Bologna / DICAM; L. Zanchi, M. Delogu, University of Florence / Department of Industrial Engineering Nowadays bio-composite materials have increased automotive market penetration, whose intend is to produce environmental friendly products while maintaining their competitive edge [1]. In particular wood-fibers is considered a more environmental friendly alternative to the synthetic reinforcements as talcum, glass and carbon fibers. The reason behind is due to their less impact on production issues (sharp reduction of toxic and fossil materials as well as for the energy expenditure, light-weight profile and more energy credit through their incineration [2-4]. This study presents the application of a new industry engineered wood-based product, called Woodforce®, for the production of an automotive PedalBox Support (PBS); in particular, the environmental advantages/disadvantages are presented and discussed when such material is thought to substitute the glass fibres. At this scope, a comparative environmental analysis has been performed between two materials - a standard material (with glass-fibres content) and an innovative (with woodchip content) - which perform the same function. 1. Reference used: Lampen A., Carus M. 2016. Wood and natural fiber composites current trend in consumer goods and automotive parts. Reinforced Plastics 60: (3) 170-173. [2] Joshi S.V., Drazal L.T., Mohanty A.K., Arora S. 2003. Are natural fiber composites environmentally superior to glass fiber reinforced composites? Composites: Part A 35 (2004) 371– 376. [3] Boland S. Claire, De Kleine R., Keoleian G.A., Lee E.C., Kim H.C. Wirtz, Research Institute gaiac / SETAC Europe 28th Annual Meeting Abstract Book

620 Potential new soil test requirements for the risk assessment of pesticides in the European Union: do we have the right methods? J. Roembke, ECT Oekotoxikologie GmbH; J. Sousa, University of Coimbra / Department of Life Sciences

In February 2017, the European Food and Safety Authority (EFSA) published a Scientific Opinion entitled “Scientific Opinion addressing the state of the science on risk assessment of plant protection products for in-soils organisms”. This document summarizes various scientific issues relevant for the risk assessment of plant protection products (PPPs) for in-soil organisms, i.e. micro-organisms and invertebrates (plants are covered in another EFSA document). The aim of this document is to provide an overview on existing regulatory requirements in soil ecotoxicology, but – more importantly – it also summarizes the scientific background for a new approach. In the future, soil risk assessment should be based on the ecosystem service approach, meaning that important services at agriculture landscapes (e.g. nutrient cycling, soil structure maintenance, just to name a few) have to be protected. In consequence, protection goals for soil organisms are to be defined. This contribution will focus on one important question, assuming that the new risk assessment approach is put in practice: Do we have the appropriate (e.g., scientifically sound, robust, standardized) methods to cover the new data requirements? In this contribution we focus on four aspects of the new methodology: Organism groups, endpoints, soils, regions (i.e. agricultural practices are not covered). In fact, there are various research needs which have to be addressed asap, i.e. before the new requirements will be put in practice. We focus our discussion on those test systems which are already standardized (or which are in the process of being standardized in the foreseeable future). Besides OECD also methods published by ISO or national organisations such as Environment Canada are considered, knowing that the latter often have to be adapted in order to cover the specific needs of PPP environmental risk assessment schemes. In addition, the same criteria as in other comparable approaches will be used and examples of suitable methods will be given. Most important is whether they can be modified in a way that they are useful under the upcoming EFSA regulations.

621 Poster spotlight: TH154, TH155, TH156

Emerging technologies and related raw materials requirements scenarios: the role of life cycle thinking

Wood-fibres composite in substitution of a synthetic material to enhance sustainability purposes for automotive sector S. Maltese, Magneti Marelli SpA / Powertrain Division; a. bonoli, DICAM- Alma Mater Studiorum - University of Bologna / DICAM; L. Zanchi, M. Delogu, University of Florence / Department of Industrial Engineering Nowadays bio-composite materials have increased automotive market penetration, whose intend is to produce environmental friendly products while maintaining their competitive edge [1]. In particular wood-fibers is considered a more environmental friendly alternative to the synthetic reinforcements as talcum, glass and carbon fibers. The reason behind is due to their less impact on production issues (sharp reduction of toxic and fossil materials as well as for the energy expenditure, light-weight profile and more energy credit through their incineration [2-4]. This study presents the application of a new industry engineered wood-based product, called Woodforce®, for the production of an automotive PedalBox Support (PBS); in particular, the environmental advantages/disadvantages are presented and discussed when such material is thought to substitute the glass fibres. At this scope, a comparative environmental analysis has been performed between two materials - a standard material (with glass-fibres content) and an innovative (with woodchip content) - which perform the same function. 1. Reference used: Lampen A., Carus M. 2016. Wood and natural fiber composites current trend in consumer goods and automotive parts. Reinforced Plastics 60: (3) 170-173. [2] Joshi S.V., Drazal L.T., Mohanty A.K., Arora S. 2003. Are natural fiber composites environmentally superior to glass fiber reinforced composites? Composites: Part A 35 (2004) 371– 376. [3] Boland S. Claire, De Kleine R., Keoleian G.A., Lee E.C., Kim H.C. Wirtz, Research Institute gaiac / SETAC Europe 28th Annual Meeting Abstract Book

623 Resource depletion of a Lithium ion battery cell technology M. Cusenza, Università degli Studi di Palermo; S. Bobba, G.A. Blengini, Politecnico di Torino; M. Cellura, M. Mistrutta, University of Palermo Lithium-ion batteries (LIBs) are the dominating storage technology for electric vehicles (EVs). Different types of LIBs, using diverse cathode materials, are typically available in the market, such as LiMnO$_2$, Li(Ni$_{1/3}$Co$_{1/3}$Mn$_{1/3}$)O$_2$. The cathodes contain a wide range of raw materials (RMs), among which e.g. cobalt is in the 2017 list of CRMs for the Europe Union (EU). CRMs are both of high economic importance for the EU, and vulnerable to supply security. In the last years, the increasing demand of LIBs has triggered a growing interest in the need to ensure the security and the sustainability of the supply of the critical RMs. The extensive use of LIBs and in general in EVs. In this context, lithium rich layered oxides from the class Li$_x$Mn$_{1-x}$O$_2$- (1-x)Li$_2$MnO$_3$ (M=Ni, Co, Mn), known as LNO – NCM, have drawn attention as cathode material due to their high discharge capacity and lower cobalt content, compared with the Ni-Co-Mn cathodes (NMC). In this context, the authors carried out a Life Cycle Assessment of an 11.4 kWh LMO-NMC battery cells usable in plug-in EVs with the view of identifying of the potential risks associated with the use of lithium rich layered oxides and their impact on the mineral, fossil and renewable resources depletion (MFRRD); to estimate the requirement of CRMs; to identify the contribution of each cell component to the MFRRD; to compare the LMO-NMC LIB cell technology with other NMC cell technologies available in the literature, with reference to the MFRRD and CRMs requirements. The LMO-NMC battery cell technology is modelled as 0.5LiMnO$_2$ - 0.5(Ni$_{1/3}$Co$_{1/3}$Mn$_{1/3}$)O$_2$ using both primary and secondary data. The cells of the 11.4 kWh LMO-NMC battery are selected as functional unit. The system boundaries include RMs supply, manufacturing, transports and infrastructures. The results show that the LMO-NMC cells have an impact on MFRRD of 0.34 kg Sb$_{eq}$ The relevant share of MFRRD (34%) is caused by the cobalt sulphate production used in the cathode. Of the 27 CRMs for the EU, the analysis shows the relevance of only two of them: cobalt and barite. From the comparison with the NMC cell, carried out with reference to 1 kWh of nominal capacity, results that the MFRRD impact and the cobalt requirement of the LMO-NMC technology is lower, respectively, of a percentage equal to -4.4% and -29% than those of the NMC. The results indicate that the LMO-NMC cell could be a suitable technology to meet the demand of the EU market as it involves a lower impact on MFRRD and a lower consumption of CRMs compared to the NMC cell.

624 Analysing the environmental impacts of alternative solutions for passenger transportation: LCA of a charging station for e-bicycles G. Rizzello, University of Rome Tre / Department of Business Studies; R. Salomone, L. Giuttari, G. Saia, G. Ioppolo, University of Messina / Department of Economics; M.C. Lucchetti, University of Rome Tre / Department of Business Studies

The transport sector causes significant environmental impacts that are mainly connected to the passenger car activities. In this context, strategies for reducing the environmental impacts related to the transport sector are required by moving from cars to alternative transport vehicles, such as electric bicycles. Although many studies have been focused on the application of the Life Cycle Assessment (LCA) method for assessing the environmental impacts of electric vehicles (EVs), there are...
few information regarding the environmental impacts connected to the life cycle of the charging infrastructures to operate the EVs, and, in particular, regarding the environmental performance of charging station for e-bicycles. This study aims to evaluate, through the application of the LCA method, the environmental burdens of a stand-alone charging station for electric bicycles manufactured in Italy, assuming its installation and utilisation in Italy with a lifetime of 15 years. The investigated stand-alone charging station is composed of eight designated positions for charging the e-bicycles' battery and of two wind-solar modular systems that are respectively equipped with one photovoltaic (PV) module and three low profile vertical axis wind turbines. The structure is also geared with a system connected to the conventional electricity grid which allows to provide energy when the wind and solar conditions are insufficient. The analysis also includes the whole life cycle of e-bicycles, assessed as the potential avoided production of conventional electricity which may be replaced by the electricity surplus produced by the wind-solar system. The functional unit (FU) adopted to perform the analysis is one charging station installed in Italy and used for a lifetime of 15 years. System boundaries follows a cradle-to-grave approach and include four main phases: 1) production phase, 2) installation phase, 3) use and maintenance phase, and 4) end of life phase. The results demonstrate that the Climate Change impact related to the whole life cycle of the investigated charging station is 13,816.5 kg CO2 eq per FU.

The main environmental impacts are due to the production phase and, in particular, are connected to the production of the e-bicycles, while higher environmental benefits are connected to the replacement of conventional electricity with the energy surplus produced by the wind-solar modular system installed on the charging station.

625 Raw materials requirements scenarios for the electric mobility penetration in the Italian urban vehicle fleet: a life cycle thinking approach combined with raw materials assessments

L. Cutaia, C. Chiaradia, P. Porta, ENEA; M. La Monica, C. Scaglirano, CINIGeo

The automotive fleet is gradually changing from internal combustion to hybrid and electric engines. This is fostered by the European policies concerning the need of reduction in pollutant emissions and in the improving of urban air quality. Currently most of the analyses carried out focus on the comparison of the emissions related to the different typologies of cars and on the CO2 emission cut generated by the consumption of electricity produced from renewable sources in the use phase of the cars. The proposed approach wants to investigate the need of raw materials in future scenarios of electric technology penetration in the urban vehicle fleet. Focusing on the Italian urban vehicle situation and considering the current technology used for the electric batteries and the recharging systems, the study account for the raw materials needed for the penetration scenarios of the electric mobility adopting a Life Cycle Thinking perspective. The authors have been beyond a mere quantification of the primary materials required for the progressive electrification of the urban fleet, evaluating the raw materials availability from a market perspective and defining the limiting factor among three basic material of the current electric technologies (lithium, graphite and manganese). Therefore this work, starting from an evaluation of the material needed for different electric mobility penetration scenarios in the Italian urban fleet, wants to highlight the market dynamics especially for 3 primary materials widely required in the electric mobility focusing also on the possibility offered by a transition to a circular economy, investigating the green mining potential available for these materials both for their recovering in the same technology and in others fields.

626 Coupling dynamic carbon accounting and partial-equilibrium econometric model for energy policy assessment

A. Alberts, P. Collet, D. Lorne, IFPP / Economics & Technology Intelligence; A. Benoitis, CIRAD / UPR BioWooELSA research group; A. Hélias, Montpellier SupAgro / LBE ELSA

Energy production and consumption is the main driver for anthropogenic GHG emissions, and in the French context, the transportation sector is the principal emitter accounting for almost one third of these emissions. The growing need to reduce GHG emissions and mitigate climate change demands tapping alternative energy resources, as differently enforced by energy policies (e.g. the French Transition for Growth Act). LCA scholars increasingly assess the environmental performance of the advance biofuels, but mainly from a static perspective. Results are therefore limited to linear simplifications, whereby long-term impacts might be neglected or underestimated. New dynamic LCA approaches have been suggested, however no consensus is available on how to treat CO2 sequestration dynamics over different timeframes. This study further addresses the temporal shortcomings of bioenergy systems while considering future outlooks and consequences on the market dynamics. The approach consists of a hybrid-approach combining the MIRET energy systems model with dynamic Chio accounting models towards dynamic LCA. The former—a prospective techno-economic partial-equilibrium model covering the French energy-transport sector—represents scenario-dependent outputs over a long timeframes (2007 to 2050), exploring optimisation options under no-policy and policy-driven constraints. The latter assesses biomass growth and allometric relations representing the Chio fixation of a vegetation species per hectare on an annual basis, and thus the time-dynamic Chio flows between the atmosphere and the technosphere. The assessed Chio flows primarily originate from lignocellulosic biomass and their co-products generated from MIRET outputs under business as usual and normative scenarios. The transformed Chio inventories are then combined with both dynamic and static LCA characterisation factors, towards a comparison of both approaches. The results show that the time factor is an essential component to properly assess long-term Chio sequestration potentials and climate benefits of lignocellulosic biofuels. The combination of technological innovation and market dynamics in a transitioning energy system expands the assessment boundaries providing insights into least cost (economic optimisation) and low carbon (Chio sequestration) options influenced by policy and decision constraints. Future refinements addressing other bioenergy paths are envisaged.

627 Poster spotlight: TH304, TH309, TH314

Developments in the use of bioassays for chemical and environmental risk assessment (II)

628 SIMONI: Smart integrated monitoring of the water quality

R. van der Oost, Waternet / Onderzoek en Advies; G. Sileno, Waternet / Research and Development; M. Thao Nguyen, Waterproof, L. Moria, Waternet / Water Systems

At present, regular water quality assessment is almost exclusively performed by target chemical analysis of substances. Scientific research over the last decades, however, demonstrated that water quality assessment with only chemical analyses is not reliable. Since over 100,000 harmful substances may be present in the aquatic environment, a paradigm shift from ‘substances’ to ‘effects’ has to be encouraged in order to implement a more holistic approach in regular monitoring. Therefore, an alternative Smart Integrated Monitoring (SIMONI) strategy has been developed by Dutch water research institutes. The purpose of this 2-tiered strategy is to obtain more reliable information on the chemical water cycle quality without increasing the monitoring costs. Key factors for generating this model were the selection of the most relevant bioassays and the design of effect-based trigger values (EBT). Tier 1 of the strategy combines micropollutant concentration by passive sampling with testing of 15 biochemical endpoints. This hazard identification makes the distinction between low, acceptable and increased ecological risks. Only at sites where tier 1 indicates increased ecological risks, a customized tier 2 research is performed to identify the chemicals that cause the bioaffective effects and to evaluate if observed in vitro effects pose a serious in vivo threat to the ecosystem. The present paper is focused on demonstrating the field feasibility of the SIMONI strategy. The strategy has been applied and validated in more than 100 Dutch field studies. Results of several of these monitoring studies will be presented in order to demonstrate the field feasibility of the SIMONI strategy for identifying hot-spots of chemical pollution. It is estimated that this approach is generally applicable for agricultural sites. In addition, increased ecological risks were also observed at receiving water receiving effluents, sewage overflows and landfill runoff. A tier 2 approach is the assessment of the mixture toxic pressure on the ecosystem by using chemical analytical results for msPAF determination (potentially affected fraction of water organisms due to multiple substance). At two sites with increased ecological risks due to micropollutants, similar classifications were obtained with bioanalytical and extensive chemical analyses. The 2 tier research at agricultural greenhouse areas identified eight pesticides contributing most to the increased environmental risks.

629 Bioassay battery responses to POCIS and Speedisk passive sampler extracts

M. de Baat, University of Amsterdam / IBED-FAME; M. Thao Nguyen, Waterproof; R. van der Oost, Waternet / Onderzoek en Advies; W. van den Berg, Waterproof Laboratory, Research and Validation; P. de Voogt, University of Amsterdam / IBED; M. Kraak, University of Amsterdam / IBED-FAME

A large portion of the toxic effects observed in surface waters cannot be explained by compounds that water authorities regularly monitor. Since chemical analysis of the myriad of substances present in the aquatic environment is practically and economically not a viable option, there is a growing interest in effect-based monitoring. Additionally, concentrations of compounds typically vary over time and therefore traditional grab sampling only provides a snapshot of the chemical make up of a water body. The effects observed in surface waters cannot be explained by compounds that water authorities regularly monitor. Since chemical analysis of the myriad of substances present in the aquatic environment is practically and economically not a viable option, there is a growing interest in effect-based monitoring. Additionally, concentrations of compounds typically vary over time and therefore traditional grab sampling only provides a snapshot of the chemical make up of a water body. The effects observed in surface waters cannot be explained by compounds that water authorities regularly monitor. Since chemical analysis of the myriad of substances present in the aquatic environment is practically and economically not a viable option, there is a growing interest in effect-based monitoring.
630 Endocrine modulation and toxic effects of sunscreen chemicals, Octocrylene and Benzophenone, on zebrafish

Q. MENG, The Chinese University of Hong Kong; K. Chan, The Chinese University of Hong Kong / Life Sciences

Sunscreen chemicals are used to prevent the skin and hair of human from the harm of sunuvioleic light, and they are widely applied in a variety of personal care products. These materials are discharged to the environment continuously as daily care products. At present, many kinds of UV filters are found in surface water and polluted water, and their concentrations in the urine and blood of human are much higher than that in the environment, indicating that the UV filters have the possibility to bioaccumulate to cause toxic effects in the accumulated organisms. Benzophenone-3 (BP3), benzophenone-1 (BP1), benzophenone-4 (BP4), octocrylene (OC) are such chemicals that have been detected in environmental samples and linked to alterations in estrogen receptor signalling pathways and oxidative stress. In this study, zebrafish larvae and a liver cell model of zebrafish liver cells, the ZFL cell line, will be used to investigate the potential risks of BPs on liver cells and the ZFL cell line, will be used to investigate the potential risks of BPs on liver cells and the ZFL cell line will be used to investigate the potential risks of BPs on liver cells as well as non-receptor mediated activity has been directed analysis, i.e. high-resolution LC-QTOF and tested on estrogens, androgenic and glucocorticoids. Also bioassay chromatograms of other endpoints will be discussed. This study demonstrates the value of toxicity profiling with bioassays as first tier in the monitoring of water quality. In case observed activities exceed trigger values, additional risk assessment is needed and the HT-EDA platform can help to characterize and ultimately identify the responsible compounds.

631 Current status of in vitro bioassay approach in environmental risk assessment of endocrine environmental mixtures and individual organic contaminants

M. Machala, Veterinary Research Institute / Chemistry and Toxicology; K. Pencíková, S. Strápacová, Veterinary Research Institute, Brno / Chemistry and Toxicology; L. Svrčková, Veterinary Research Institute, Brno / Chemistry and Toxicology; J. Neca, M. Ciganek, Veterinary Research Institute, Brno / Chemistry and Toxicology; I. Bartonková, Z. Dvorák, Faculty of Science, Palacky University, Olomouc; J. Topinka, Institute of Experimental Medicine, CAS, Prague; J. Vondrácek, Institute of Biophysics, CAS, Brno

Since 2001, our laboratory has continuously employed an ever growing set of in vitro bioassays combined with a detailed chromatographic analysis, and in some cases the effect-directed analysis, in order to identify principle modes of action of contaminants bound to river sediment, airborne or diesel exhaust particles. Toxicity profiling of selected individual contaminants was used as a complementary approach, which aimed to identify major toxic modes of action and principle contributors to specific toxicity effects. The AhR-mediated activity has been recognized to play a key role in toxicity of organic extracts of abiotic environmental mixtures. Using the DR-CALUX assay, we established relative effective potencies (REPs) for a large variety of poly cyclic aromatic compounds, including PAHs, methyl-, nitro- and oxy-PAHs, thiophenes, dithiophuranis, benzo[c]phenanthrene and carbazoles, which complemented the available data on polychlorinated biphenyls, dibenzo-p-dioxins and dibenzofurans. Recently, we also developed REP values in human AhR-dependent reporter gene assays, in order to compare the potencies in rodent and human models. Using a set of various CALUX assays, receptors and selected individual classes of organic contaminants. The general outline of those studies will be illustrated using an example of a detailed in vitro toxicological evaluation of standard reference material of diesel exhaust particles (SRM 2975), with an aim to document both the complexity of the observed effects and the difficulties faced when applying these data in risk assessment of complex mixture. [The study was supported by the Czech Science Foundation, grant no. P503-12-G147.]

632 Hormone-like activities in waste water characterized by CALUX bioassays, Chromatographic analysis and Effect-directed Analysis

Y. van Oorschot, R. ten Broek, The Water Laboratory; M. Lamoree, VU University, Department Environment & Health / Department Environment & Health; N. Zwart, VU University Department Environment & Health; C.J. Houtman, The Water Laboratory

Emission of compounds with biological activities from waste water treatment plant (WWTP) into the environment is a topic of concern to ecology and drinking water companies. We investigated the occurrence of hormone-like activities in WWTP samples and pursued to identify compounds responsible for them. To this aim, CALUX bioassays and a UPLC-Q-MS target analysis method for hormones used in high volumes in pharmacy were applied. In addition, a novel high throughput Effect-directed Analysis (HT-EDA) platform was used to separate compounds in the extracts with high resolution LC-fractionation creating 288 4.79 sec.-fractions that were tested in the bioassays. In parallel, QToF high resolution MS data were recorded to correlate compound identity to peaks from the 'bioassay chromatograms' reconstructed from the bioassayed fractions. All five types of activities tested were observed in the WWTP samples. Androgenic and estrogenic activities were almost completely removed during WW treatment, anti-androgenic activity was not removed as completely. Androgenic and progestagenic activities were present in similar concentrations in untreated as in treated WW. The glucocorticoid activity in influent was mainly predicated by prednicarbact, trimcinolone acetounde, dexamethasone and amiconidine. In effluent however, detected hormones could only explain 15% of the activity, indicating the presence of unknown (metabolites of?) glucocorticoids in effluent. The androgenic activity in effluent was almost fully removed and the progestagenic activities were almost completely removed during WW treatment. The anti-androgenic and progestagenic activities were possibly contributed to the presence of aldosterone and androstenedione and testosterone. Application of the HT-EDA platform-delivered bioassay chromatograms of the WWTP effluent in which active compounds were separated into sharp peaks. The glucocorticoid activity appeared to be caused by at least four different compounds (peaks), not being dexamethasone. The androgenic activity was fractionated into a small peak probably attributable to co-elution of testosterone and androstenedione. The QToF MS data are currently interpreted to elucidate the identity of the unknown glucocorticoids. Also bioassay chromatograms of other endpoints will be discussed. This study demonstrates the value of toxicity profiling with bioassays as first tier in the monitoring of water quality. In case observed activities exceed trigger values, additional risk assessment is needed and the HT-EDA platform can help to characterize and ultimately identify the responsible compounds.

633 Non-target screening and identification of emerging pharmaceuticals and their transformation products in wastewaters

c.g. Pagés, University of Bordeaux / EDCA / LPTC / UMR 5805 CNRS; M. Dévérié, University of Bordeaux / EPOC / LPTC / UMR 5805 CNRS; E. Mailtot-Maréchal, INERIS / UMR SEBIO ECOT; E. Geneste, University of Bordeaux / EPOC / LPTC / UMR 5805 CNRS; S. Ait-Aissa, INERIS / UMR SEBIO ECOT; H. Budzinski, University of Bordeaux

Wastewaters represent a major pathway of introduction of EDCs into the aquatic environment. Considering the large diversity of compounds that might be introduced into wastewater, many EDCs are currently unknown. Therefore, the characterization of the presence and identification of EDCs in wastewater are major issues in order to assess their occurrence in natural waters and the associated risks for wildlife. To date, in vitro assays based on luciferase reporter gene expression, are available to assess the biological activities of samples in a quantitative, sensitive, specific and fast way. The comparison of concentrations derived from bioassays and from chemical analyses allows assessing the contribution of micropollutants to the overall biological or toxic activity of a sample. In this study, a systematic approach combining effect-directed analysis (EDA) and high resolution spectrometry was applied to several urban WWTPs to establish an overall contamination diagnostic and to identify major contaminant that could be released in environment. For this purpose, crude extract from influent and effluent of an urban WWTP were analyzed by LC-QTOF and tested on estrogens, androgens and glucocorticoids receptors. The non-target screening allowing detection more than 7000 and 4000 signals in influent and effluent extracts of WWTP, respectively. We observed that 70% of compounds detected in effluent were introduced by the treatment process suggesting the formation of transformation products. Concerning the strategy implemented to identify compounds of interest, we was decided to use effect-directed analyses methodology to have a tool to target active compounds in relation with selected biological activity. To this end, the crude extracts were fractionated by HPLC and biologically active fractions were isolated for further chemical identification. Biological fractionation profile of samples indicated the presence of estrogenic and glucocorticoid activities at all studied sites with very similar fractionation patterns between sites, highlighting major and recurrent individual fractions. The identification of active compounds was performed using LC-QTOF and several drugs and their transformation.
products (e.g. O-desmethyl venlafaxine, O-desmethyl tramadol) were recurrent found. This approach combined to LC-QTOF has allowed establishing a list of systematic detected non-target compounds in several wastewaters.

Indigeneity and Science: A collaborative work in progress

634 The Nechako White Sturgeon Recovery Initiative: A discussion of species at risk conservation, scientific outreach, community and First Nations support

T. Lane, University of Saskatchewan; C. Williamson, Freshwater Fisheries Society of British Columbia; S. Shah, University of Saskatchewan / Toxicology Centre; M. Hecker, University of Saskatchewan / School of the Environment & Sustainability and Toxicology Centre

Nechako white sturgeon (Acipenser transmontanus) are a genetically unique population of fish which have inhabited the Nechako watershed for roughly 10,000 years. Within the last 50 years this population has suffered significantly because of anthropogenic activities; the Status of the Convention of International Migratory Waterfowl in Canada identified the Nechako white sturgeon as a Nationally Significant Population. In 2006, this population was further listed as endangered under the Species at Risk Act. Prior to both designations in 2000, the Nechako White Sturgeon Recovery Initiative (NWSRI) was established in Vanderhoof, British Columbia. The goal of the initiative is the conservation and recovery of this one-of-a-kind population of white sturgeon which hold intangible cultural value within communities surrounding the Nechako watershed. A 5-million-dollar aquaculture facility, the Nechako White Sturgeon Conservation Centre was designed specifically for the NWSRI, built, and opened in 2014 to provide the resources to further support this conservation effort. This facility was a product of over a decade of work by a Technical Working Group (TWG) and a Community Working Group (CWG). The TWG includes biologists, industry and First Nations members who have a vast knowledge of white sturgeon. The goal of the TWG is to use the best available science, local and traditional knowledge to determine why the Nechako white sturgeon population is declining and to develop a plan to rehabilitate this population of fishes. The CWG is composed of First Nations members, local and regional governments, industry, and public volunteers. The CWG plays a vital role in communication, public outreach, and promoting community involvement. Activities of TWG and CWG support the mandate of the NWSRI through direct involvement of First Nation communities, volunteers and students. The Emergency Sturgeon Live Release Boat Kit program is an example of multiple First Nation communities working in union with the NWSRI to promote conservation and stewardship of Nechako white sturgeon. The annual Juvenile Sturgeon Release event involves students, volunteers and First Nations who release thousands of juvenile sturgeons, which were reared from eggs by the TWG, back into the Nechako watershed. The NWSRI is a unique conservation effort that promotes and utilizes the support of scientists, First Nations, volunteers and students to engage the community in the conservation of an endangered species.

635 The NSERC CREATE H2O Program on First Nations Water and Sanitation Security: Case Studies on Drinking Water Quality

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The NSERC CREATE H2O program is the first science-engineering research training program in Canada that combines technical water and wastewater management training with Indigenous theory, law and methodological skills training. Since its inaugural year in 2013, the program has trained 86 students who have collectively worked with 30 First Nations communities in the provinces of Manitoba, Ontario and Saskatchewan, Canada. 33% of the university students and postdocs enrolled in the program self-identify as Indigenous. This presentation provides an overview of the approaches the program is using for: engaging communities and students in research training activities, Indigenousizing science and engineering curricula, and designing advocacy strategies to support clean drinking water as a human right in First Nations communities in Canada. Case studies are presented to demonstrate the community-based monitoring programs implemented to examine drinking water quality in First Nations homes. First Nations communities participating in the research have various types of water distribution systems. Overall, water samples were collected from: lakes and groundwater (source water); water treatment plants, water trucks and community standpipes; homes relying on piped water, wells, above-ground cisterns and underground cisterns; and buckets/drums in homes without running water. Water analysis included standard measures of chemical and bacterial parameters, DNA and RNA techniques for microbial profiling, and the quantification of antibiotic-resistance genes in water samples. The main findings are that despite effective water treatment plants in communities, the tap water in many First Nations homes contained fecal bacteria at alarmingly high levels and antibiotic-resistance genes were also detected in a range of drinking water samples. The issue of poor drinking water quality in First Nations communities in Canada remains unsolved and there is an urgent need for improved monitoring and upgrading of infrastructure, especially in communities relying heavily on cisterns and community standpipes for drinking water supplies. Most importantly, investments to connect homes directly to water treatment plants via improved pipelines is key to reducing human exposure to waterborne illnesses, while enhancing options for families to participate in economic development, food security and spiritual and cultural wellbeing in their communities.

636 Rare earth elements (REEs) in the Canadian Subarctic: scientific perspectives and community engagement with environmental monitoring in Nunavik, Northern Quebec

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Many communities in Canada’s North are increasingly concerned about the impacts of large-scale socio-environmental changes, such as climate change and insecure on our lives, water and communities. In Nunavik, Northern Quebec, where Indigenous communities is the environmental impact of mining, including the large number of rare earth element mining projects that are currently being developed in northern Canada. Rare earth elements (REEs) are a chernically-group of contaminants of emerging concern, which includes the 15 trivalent lanthanide metals. Not particularly rare, REEs are increasingly exploited for essential uses in high-tech industries, including electronics, clean energy, and agriculture. Although knowledge of the fate of REEs and their impact on natural ecosystems is critically needed as emissions increase, few ecotoxicological studies exist for REEs particularly field studies on their bioaccumulation and food web dynamics. To investigate how REEs behave in remote northern food webs, samples were collected from community-driven collaborative research projects with the Inuit communities of Kuujjuaarapik-Whapmagoojistiu (K-W) and Kangiqsulujuaq in Nunavik (Northern Québec). The combined objectives of these projects were a) to study the behaviour of REEs in northern ecosystems before the start of mining activities and b) to engage community members in the research process through the co-development of objectives, sample collection and the sharing of research results. We will share perspectives from our communities and community engagement activities. Can we detect REEs in freshwater, marine and terrestrial plants and animals? Do REEs bioaccumulate and biomagnify in northern food webs? How to best establish a sustainable community-based environmental monitoring program? Can we engage the youth in environmental monitoring and science education? Wildlife harvesting and tissue sampling was partly conducted by local hunters, including a range of key species important for both biomonitoring and country food. Indigenous knowledge was used in the study design, to coordinate local sampling, and to decide when, where and what species to collect. Our study presents novel data on the behaviour of REEs in northern ecosystems and recommendations for establishing sustainable and effective community-based environmental monitoring projects with Indigenous communities for emerging contaminants.

637 Te Ohu Mō Papatūānuku: A Collective Response to Healing

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The use of pentachlorophenol (PCP) an anti-rotant in timber treatment, with subsequent disposal of chemically treated wood waste in the Whakatane District of New Zealand has resulted in a legacy of contamination. There are 36 identified wood waste sites located on private and public lands, as well as the customary lands and waters of the indigenous Ngati Awa people. The pervasive effects of contaminants upon both human and environmental health has led to the formation of multiple Indigenous groups. The Te Ohu Mō Papatūānuku: A Collective Response to Healing project is supported by indigenous members of Ngati Awa, whilst being strongly supported by scientists, local government agencies, and industry. As a consequence of ongoing research, the use of a rather unique approach utilising combined myco- and phytoremediation to remediate dioxin-contaminated land has been adopted. Whilst implementation of the approach is underpinned by science, the use of “nature to heal nature” is an approach that resonates with the indigenous community. Contemporary environmental problems resulting from anthropogenic activities often require the use of scientific based solutions. Hence, even when indigenous participation is encouraged by the scientific community as part of the problem-solving process, the contribution of indigenous knowledge may be considered of less value than scientific knowledge. Of vital importance to ongoing environmental remediation, however, is the role of indigenous knowing — indigenous relationships informed by binding and enduring familial links with lands, waters, and people. This presentation builds upon previous presentations detailing the journey of the Te Ohu Mo Papatuanuku research collaboration — using a synthesized approach —
Drinking water advisory levels have been adopted by many regulatory agencies to reduce chronic exposure to persistent fluorochemicals. However, most U.S. advisory levels are based on the assumption of approximately 20% of overall PFAS intake comes from drinking water. Better characterizing the relative importance of drinking water to overall human exposures is important for developing health protective guidelines. Most previous investigations that have associated drinking water PFAS exposures and total body burdens of these compounds have focused on highly contaminated sites. Exposure to PFASs for the general population of individuals from geographically diverse areas is thus less understood. Here we investigate the relative importance of drinking water for total PFAS exposure among women in the Nurses’ Health Study (NHS), a large U.S. based cohort study. Our analysis included U.S. women who provided a residential drinking water sample in 1989-1990. We measured concentrations of 11 PFASs in a subset (n=111) of archived drinking water samples and serum samples. We evaluated the relative importance of home tap water for measured levels of PFASs in human using both statistical and toxicokinetics (TK) models. Results suggest that home tap water is a significant exposure source for general American women. In 1989-1990, the median contribution of drinking water to serum PFASs in women in the NHS cohort was 8.8% to 39% for the five PFASs modelled. This ratio varies across individuals and is potentially much higher by up to a factor of 2-3. We will investigate how this ratio varies geospatially and whether it is associated with distance to well-known point sources. The spatial analysis results will also be discussed in the presentation. By comparing PFAS concentrations in archived tap water sample with recent tap water samples matched on the township, we found the fraction of quantifiable PFASs (i.e. known PFASs) has decreased in most towns and unknown extractable organic fluorine (EOF) has increased. Our analysis suggests tap water may be a significant exposure source for five PFASs among a group of U.S. women from diverse geographical areas. Increases in unquantified EOF in recent tap water suggest additional quantification would be worthwhile. Other exposure sources such as consumer products are suspected to dominate overall exposure of individuals in the NHS cohort prior to the restrictions and regulations of legacy PFASs in the U.S.

Consultation with Remote, Indigenous Communities in the Northern Territory, Australia

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On 18 September 2016 the Northern Territory (NT) Government announced a scientific inquiry into hydraulic fracturing of onshore unconventional reservoirs in the Northern Territory (the Inquiry) under the Inquiries Act (NT). The Inquiry was required to assess the available scientific evidence to determine the likely nature and extent of the environmental impacts and risks, including cumulative impacts and risks, associated with hydraulic fracturing of unconventional reservoirs and the Associated Activities in the NT. The Inquiry was specifically instructed to assess whether or not there would be unacceptable impacts or risks to economic, cultural and social conditions, by developing and implementing a stakeholder engagement program. Indigenous people make up most of the resident populations in the areas of the shale-gas basins in the Northern Territory. Indigenous people are linked with their land (including waterbodies) by their ancient traditions and continue to use their land in accordance with those traditions. As a community, they must be able to maintain their cultural traditions relating to that land in order that their ownership rights continue to be recognised, from one generation to the next. Indigenous communities are therefore particularly vulnerable to degradation of the landscape and the natural systems it supports. Therefore, the Inquiry undertook focused stakeholder consultation with remote indigenous communities. The process involved substantial engagement with most. The community feedback gained thereby was a vital input into the Inquiry’s assessment of the potential impacts of shale gas development in the NT.

Incorporating cultural values and perspectives of First Peoples’ (Aboriginal) People into water planning, science and environmental water management

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Australia is the oldest inhabited continent on Earth, yet is has been the traditional lands of its original inhabitants Australia’s First Peoples for thousands of generations. Protecting water landscapes (surface and ground water) has always been a high priority for survival in a dry landscape, and protecting water remains a cultural obligation. The challenge for First Peoples is to ensure their value and relationship with water is not diminished or excluded by modern day water planning and science or from environmental flow management. First Peoples acquire the right wisdom and traditional science and knowledge and many indicate that their worldview is seeing water as inseparable from the land and the sky, bound by traditional lore and customs for its protection. For Australia’s First Peoples, occupying an ever drying landscape, traditional knowledge of finding and protecting water sites has been integral to their survival for so long. This paper will explore relationships between First Peoples and water planning and environmental water management in three ways. Firstly, history, challenges and institutional responses in integrating First Peoples cultural values into water planning, science and management. Secondly, propose a series of on-the-ground applications of cultural water and environmental water empirically. Finally, integrating First Peoples’ science into water management will be assessed through comparisons between the Australian situation through case studies looking at models and methodologies.

Improvements in environmental exposure assessment: Development and application of tools across industry sectors, regulatory agencies, and international boundaries (II)

Tap water intake of poly- and perfluoralkyl substances (PFASs) in relation to serum concentrations in a nationwide prospective cohort of U.S. women

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Drinking water advisory levels have been adopted by many regulatory agencies to
Ecotoxicology of micro and nanoplastics: Mechanistic

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approaches to understand their risk for the environment and human health

646 Wastewater-based microplastics: Presence in wastewater effluent and effects on freshwater organisms

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Microplastic pollution has become a serious environmental concern. Microplastics associated with wastewater treatment plant (WWTP) effluent have been found globally in marine and freshwater environments. Nevertheless, the concentration of microplastics discharged via wastewater after different treatment processes remains less understood. In this study, we identified microplastics in the wastewater effluent from three major WWTPs utilizing primary, secondary and tertiary treatment processes in Sydney, Australia. A novel validated sampler was designed for in situ collection of microplastics from wastewater effluent. The sampling method was combined with an efficient sample processing procedure to enhance the accurate detection of microplastics. The results indicated that primary effluent contained an average 1.5 microplastics/L. The amount of microplastics reduced to 0.6 microplastics/L after secondary treatment and 0.2 microplastics/L after tertiary treatment. Polyester fibres and polyethylene beads were predominantly detected in wastewater effluent, which possibly originate from synthetic clothing and cosmetic products, respectively. This suggests that WWTPs can act as a significant pathway to release microplastics to the aquatic environment, given the large volume of treated wastewater being discharged on daily basis. The effects associated with wastewater-based microplastics (e.g. fibres and beads) were thus investigated by exposing four freshwater organisms, a water flea (Daphnia magna) and a sediment-dwelling midge larva (Chironomus tepperi), to microplastics in water and sediment, respectively, at concentrations within the range of environmentally realistic concentrations. A dose-dependent effect was observed after acute and chronic exposure of C. dubia to microplastics, with fibres showing more significant effects, though chronic effects were only observed at concentrations six times higher than reported environmental levels. Further, exposure to an environmentally relevant concentration of microplastics adversely affected the survival, growth, and emergence of C. tepperi. Size-dependent effects were observed with microplastics, with beads in the size range of 10-27 µm showing more pronounced effects. Our study demonstrates that microplastics are released into the environment by WWTPs and can have effects on freshwater organisms at concentrations within an order of magnitude of environmentally relevant levels.

647 What is in our plastic? In vitro toxicity of extracts from plastic products

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The ubiquitous abundance of plastic litter in aquatic ecosystems causes concern over their potential ecological and health impacts. So far, most toxicity studies focus on physical effects of plastic particles and chemical effects of sorbed environmental pollutants. The effects of chemicals intrinsically present in plastics, i.e. plastic additives and side products, attract less attention. Nevertheless, it is well established that these chemicals migrate from consumer products, thus representing a source of exposure to wildlife and humans. The aim of the current study is to investigate the in vitro toxicity of chemicals leaching from various plastic products and to characterize them using non-target chemical analysis. Different plastic types shall be ranked according to the toxicity of their leachates. Thirty-four plastic consumer products made of high-density polyethylene (HD/LDPE), polypropylene (PS), polyethylene (PP), polyethylene terephthalate (PET), polyvinyl chloride (PVC), polyurethane (PUR) or polyactic acid (PLA) were cut and extracted in a sonication-assisted, solvent-based procedure mimicking worst-case leaching. The extracts were tested for unspecific toxicity in the Microtox assay, for estrogenic and anti-androgenic activity in yeast-based reporter gene assays as well as for their potential to induce an oxidative stress response in the ARE32 assay. Two-thirds of the products leached chemicals triggering unspecific toxicity and one third of the samples induced an oxidative stress response. Nine plastic products released chemicals that were antiandrogenic and four slightly estrogenic. Overall, PVC and PUR extracts induced the highest toxicity in terms of potency and number of affected endpoints as almost all extracts triggered a high toxicity, oxidative stress and antiestrogenic activity. In contrast, PET extracts were less toxic with only one sample inducing oxidative stress. Interestingly, all PLA extracts produced a very high unspecific toxicity. Effects of HDPE, LDPE, PP and PS extracts strongly depended on the product. Our findings indicate that extracts of plastic products induce a range of toxicological endpoints, including unspecific toxicity, oxidative stress and endocrine activity. Screening different plastic types consistently identified higher levels of toxicity and different toxicological response. This demonstrates that a substantial part of plastic products are a potential source of exposure to toxic chemicals.

648 Microplastic size-dependent toxicity, oxidative stress induction, and multixenobiotic resistance (MXR) inhibition in the monogonont rotifer (Brachionus koreanaus)

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Microplastics have become a great concern in these days due to their world-wide distribution, persistence, and increasing amount of small-sized plastic products due to degradation of larger plastic debris. However, little is known about their impacts on marine organisms, particularly at the molecular level. Here the dependence of microplastic toxicity to the monogonont rotifer (Brachionus koreanaus) on particle size was investigated by studying the ingestion and egestion of microplastics. Particle size (0.05, 0.5, and 6 µm) polystyrene microbeads were exposed to polystyrene microbeads led to significant size-dependent negative effects on growth rate, reproduction, and lifespan. In contrast, transmission electron microscope (TEM) analysis have revealed cellular damages in the rotifer B. koreanaus exposed to 0.05 µm microbeads, indicating nano-sized microbeads would cause more serious impacts on aquatic organisms. To further explore the defense mechanisms in response to different sizes of microbeads, the activities of several antioxidant-related enzymes and phosphorylation statuses of mitogen-activated protein kinases (MAPKs) were determined. Particularly, 0.05 µm microbeads have inhibited multixenobiotic resistance (MXR), resulting increase of sensitivity of rotifers to environmental pollutants. Our study provides a better understanding of molecular responses in the rotifer B. koreanaus in response to microplastics and their potential impacts on the aquatic ecosystem.

649 Sorption of model pollutants on microplastics and toxicity assessment using early life stage of zebrafish (Danio rerio)

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The growing production of plastics increased the amount of plastic debris in aquatic ecosystems. Their degradation lead to the emission of microplastics (MPs) when their size is between 1-5000 µm. MPs can result from runoff and degradation (biodegradation or weathering breakdown) of macroplastics or from microplastics and be ingested directly by organisms, causing chronic physical and/or toxicological effects. While accumulation of MPs in aquatic ecosystems is a growing concern in society, the toxicity of MPs for wildlife and the processes of sorption of organic pollutants onto MPs is very complex and poorly understood. Therefore, objectives of the present study were to investigate the sorption kinetics of two models pollutants to LDPE (Low Density PolyEthylene) microparticles for 3 months, and the toxicity linked to pollutants sorbed on microplastics, using zebrafish embryos and larvae. Results of the sorption experiment showed that a longer exposure time did not affect the sorption rate of PFOA, but affects BaP sorption to the particles. The sorption of BaP was increasing during the first 7 days, while for the remaining 3 months of the experiment, the concentration of BaP absorbed by the MPs was constantly decreased. On the toxicity aspects, no acute toxic effects were observed, using FET test with microplastics particles (vigin or spiked). No differences were found regarding sub-lethal endpoints (genotoxicity, phenotypic effect, response (PERS) and EROD activity) between the control group and fish exposed to virgin MPs, spiked MPs or computer芦ulated. Results showed that MPs can be vectors of pollutants which sorbed at the surface over time. The toxicity of MPs as carriers of POPs cannot be demonstrated with 96h of exposure to LDPE microparicles. Experiments are currently being done to test the toxicity of MPs.

650 Comparative role of microalgae and microplastics in the effects of chlorpyrifos on molecular biomarkers in marine mussels

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Plastic particles within the microns range (microplastics, MP) are increasingly present in marine ecosystems. One of the most concerning aspects of MP in marine
natural toxins and harmful algal blooms (HABs): water and food safety, analysis, toxicity, and risks (II)

Dissipation of the carcinogenic ptaquiloside in water resources
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Ptaquiloside (PTA) is a natural carcinogen found in a number of ferns Worldwide. The distribution and occurrence of PTA is best known from the Bracken ferns (genus Pteridium) which are classified in Group 2B Possibly carcinogenic to humans by WHO/IARC. The content of PTA in Bracken is highly variable (up to 5 w-%). PTA is readily leached from Bracken stands from where it can enter the soil, waterways or potentially contaminate groundwater. Several records of PTA contamination of surface water and upper groundwater exist from Denmark and Great Britain. The fate of PTA in surface and ground water has not been studied. Under sterile conditions, dissipation (= hydrolysis) of PTA in aqueous solution follows classical first order kinetics: \[ k_{\text{ext}} = k_{[H]} + k_{\text{acid}} + k_{\text{alkaline}}[\text{H}]. \]
The rate constants are: \[ k_{\text{ext}} \approx 25.7 \text{kJ m}^{-1} \text{mol}^{-1}; k_{\text{acid}} \approx 9.5 \times 10^{-3} \text{M}^{-1} \text{s}^{-1} \] and \[ k_{\text{alkaline}} \approx 4.8 \times 10^{-3} \text{M}^{-1} \text{s}^{-1}. \] The activation energy for PTA hydrolysis at pH 4.6 is approx. 75 kJ mol\(^{-1}\). Hence, hydrolysis is a function of both pH and temperature. The purpose of this investigation was to study the degradation of PTA under near-natural conditions using 10 different surface and groundwaters from Denmark and to compare the degradation kinetics with the existing model for hydrolysis.

Dissipation was fast in natural non-sterilised lake waters with half-lives from 5 to 25 h. All PTA were degraded within 200 h. Sterile controls had no degradation. Winter samples exhibited slower degradation (half-lives up to 100 h). Sterile samples followed the existing model for hydrolysis closely, i.e. no degradation at neutral pH. PTA persist considerably longer in groundwater. Half-lives in groundwater ranged from 7 to approx. 50 days with fastest degradation in alkaline waters. The existing model for hydrolysis could generally predict the rate of degradation in groundwater. However, the model did not perform well at low or high pH. Pterosin B was formed as the end product by hydrolysis in the tested groundwaters at a molar ratio of approx. 1:1 for PTA:Pterosin B. However, under weak acid conditions, only approx. 10% of the potential pterosin B were formed. The risk of leaching from Bracken stands and contamination of surface waters is highest during the winter season. PTA has the potential of contaminating mainly shallow aquifers as the compound can persist for up to several months in contact with groundwater. The risk is expected to be highest at near-neutral conditions in aquifers.
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The Mediterranean region is one of the most densely populated and industrially developed area in Spain. As a consequence, most of the rivers in this region are impacted by multiple anthropogenic stressors. One of these rivers is the Ter River (NE Spain), where human pressures are diverse and have increased in the last decades because farming, urban development and industry practices depends on its water. In addition, fluctuations in water discharge due to the Mediterranean climate create high variability of conditions along the Ter River. One of the major problems detected in Ter River in the recent years is the appearance of geosmin. This is a metabolite generated mainly by cyanobacteria and actinomycetes that, when die, is released into the water, giving it a bad smell and taste. Although some studies have described that the production of this metabolite depends on environmental conditions, the factors associated with its production are still not clear. This supports an economic Sanità ActiVego from supply companies, since they cannot predict its appearance and have to act when customers complain.

The aim of this study is to evaluate the co-relation between physicochemical parameters and geosmin appearance along the Ter River during one year, and to study seasonal variability of geosmin concentration. The study has been performed in four sampling sites across the upper-middle part of the Ter basin. The sampling framework varied throughout the year, with the potential to evaluate the geosmin concentration in different periods (February to June, sampling was performed weekly from June to December, sampling was performed monthly. The parameters analysed have been nutrient concentration, suspended solids, organic material, turbidity and geosmin concentration in water. Biofilm samples were taken in order to analyse the chlorophyll a content. The results obtained clearly reflected the seasonal variation in the concentration of cyanobacteria being its concentration higher in winter (32 ng/L). They also evidenced the N/P ratio as one of the key factors involved in the geosmin formation. However, a more in-depth analysis of the N/P ratio in water is still necessary in order to explain the mechanisms that generate the geosmin formation within the organism. For this reason, a mesocosms experiment that tests the influence of the N/P ratio on the geosmin formation within the biofilm could be the next step to follow.

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Italian guidelines to assess and manage the risk associated to cyanobacteria blooms in water during bathing and recreational activities

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Many species of cyanobacteria thrive in different aquatic environments, where they can produce cyanotoxins with different toxicological profile. The still growing anthropic pressure and climate changes are causing the expansion in terms of time and space of their blooms, increasing the concerns for human health in several exposure scenarios. The Italian guidelines for the management of cyanobacterial blooms in bathing water, firstly drew up for the implementation of European bathing water directive (Directive 2006/7/EC), have been recently updated. A risk-based approach has been developed after a thorough revision of the current scientific knowledge on cyanobacteria distribution in the Italian Lakes and on chlorophyll and cyanotoxins biomonitoring carried out in previous seasons. The possible exposure scenarios have been considered: oral, dermal and inhalation exposure to cyanotoxins, during recreational activities, have been individually examined, to develop a framework of thresholds and actions aimed at preventing harmful effects for bathers. Three phases of attention relative to monitoring plans have been consequently defined: routine, alert and emergency, suggesting the actions to take at any moment. Parallel to environmental monitoring, a multi-step health-surveillance system has been proposed, aimed at collecting important epidemiological information and at limiting unnecessary accesses to the hospital through a screening action by local workers (lifeguards, local health units, pharmacists, etc.). All the technical/practical information on strategies, methods and tools to implement these surveillance have been foreseen in the guidelines. From the sampling of different matrices, to the analysis and the reporting to health authorities are provided. IN summary, guidelines, also by comparing international guidance values and/or guidelines, provide criteria to plan environmental monitoring activities, health surveillance and public communication systems. Finally the still important scientific gaps and research needs are highlighted.

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Identification and prioritization of emerging risks for food safety: climate change as a driver

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According to the General Food Law, the European Food Safety Authority (EFSA) is required to identify emerging risks in the fields within its mission. EFSA has developed a methodological framework for identification of emerging risk, starting from a preliminary identification of priority emerging issues through knowledge networking activities. The long term anticipation of emerging risks includes the identification of drivers. Drivers are the underlying natural or human-induced factors that directly or indirectly cause emerging risks. Climate change is recognised as a critical driver and its impact on the occurrence and toxicity producing phytoplankton, bacteria and pathogenic viruses and on other food safety domains was demonstrated. With the aim of further exploring tools to identify and prioritise emerging risks, EFSA initiated a project focusing on climate change as a driver of emerging risks for food and feed safety, including plant and animal health. A knowledge discussion group involving the major institutions involved with climate change has been created. The group will define criteria to identify relevant subdrivers (eg. rising and more fluctuating temperatures, changing precipitation patterns, increase in natural disasters etc), the issues relevant to different food safety domains including plant health and animal health, and to develop a harmonised and transparent scoring system applicable to the identified emerging issues in order to prioritise future research and risk assessment activities.

Advances in evaluating and regulating endocrine disruptors

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Hazard identification of endocrine disrupting properties of pesticides on non-target organisms: state of the art and future perspectives

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According to the Regulation (EU) no 283/2013, setting out the data requirements for active substances, in accordance with Regulation (EC) No 1107/2009 concerning the placing of plant protection products on the market, the endocrine disrupting properties of pesticides should always be assessed, as substance identified as an endocrine disruptor should not be approved. Most of the current knowledge about endocrine disruption is related to EATS (Estrogen, Androgen, Thyroid and Steroidogenesis) modalities in vertebrate organisms i.e. there is a good mechanistic understanding on how those modalities can lead to an adverse effects. The OECD conceptual framework and the OECD 150 provide an overview of the existing assays for the endocrine disruptors (ED) hazard identification and guidance on how to interpret the results of those tests. A suitable testing strategy which allows the identification of ED properties of pesticides through EATS modalities is available for some taxa of non-target organisms (i.e fish and amphibians). The analysis confirmed that the available test methods and knowledge on birds’ endocrinology do not allow a full ED assessment although they can provide supportive information. In the case of reptiles, appropriate standard test methods are completely missing. In some circumstances, extrapolation between taxa could be scientifically supported. However, consideration should be given to the taxon-specific differences. Extrapolation between mammals and amphibians regarding the ED hazard identification through the thyroidal modality has been investigated in the past. A similar analyses has not been done for EAS modalities, however, in some cases extrapolation among oviparous vertebrates is scientifically justified e.g. in the case of the steroidogenesis pathway leading to reproductive dysfunction (see more specific section). The main scope of this work is to present a critical overview of the available standardised test methodology for the ED assessment of non-target organisms, including consideration on the extrapolation between taxa. In addition, possible future perspective and research needs are discussed.

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Contaminants of emerging concern in the North American Great Lakes: Evidence of reproductive disruption from field and laboratory studies

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Analysis of nearly 500 surface water samples collected as part of the Great Lakes Restoration Initiative at 54 sites in tributaries of the North American Great Lakes Watershed confirmed the ubiquitous presence of Contaminants of Emerging Concerns (CECs) in anthropogenically impacted aquatic environments. Cluster analyses of commonly detected CECs in this data matrix suggests that the co-occurrence of approximately half of the CECs can be attributed to dichothomes urban or agricultural upstream land use. Mixtures found in watersheds with urban influences commonly contained steroid estrogens, BPA, alklyphenols, pharmaceuticals and personal care products. Agriculturally influenced sampling sites contained herbicides and pesticides in addition to BPA and alklyphenols, but mostly lacked pharmaceuticals and personal care products. Almost 3,000 resident and caged sunfish (Lepomis spp.) were collected from 27 of the 54 sampling sites and analyzed for indicators of stress associated with CEC exposure. In the presence of high aqueous CEC concentrations, glucose concentrations spiked in sunfish
plasma and liver cells exhibited toxic stress response. Canonical correspondence analyses revealed that concurrent with indicators of toxic stress, biomarkers of reproductive potential declined. To further examine the population level consequences, fathead minnows (Pimephales promelas) were exposed in the laboratory for three generations to the empirically derived urban CEC mixture at three environmentally relevant concentrations. Mixtures at environmentally manipulated concentrations enhanced fecundity in the F2 and 3 generations, while higher mixture concentrations resulted in declining fecundity. Taken together, this integrated series of studies indicates that CECs in Great Lakes tributaries may impact fish population health and sustainability.

660 AOP-informed assessment of Endocrine Disruption in freshwater crustaceans K. Tollefsen, NIVA / Ecotoxicology and Risk Assessment; M. Cronin, Liverpool John Moores University; J. Sylte, The Arctic University of Norway; F. Falciani, University of Liverpool; T. Iguchi, YOKohama City University / Molecular Environmental Endocrinology; C. La Lone, U.S. EPA / Mid Continent Ecology Division; Y. Li, NIVA Norwegian Institute for Water Research / Marine Environmental Chemistry; L. Weltje, University of Southern Denmark / Biology; L. Weltje, University of Southern Denmark / Biology; L. Weltje, University of Southern Denmark / Biology; L. Weltje, University of Southern Denmark / Biology; L. Weltje, University of Southern Denmark / Biology; L. Weltje, University of Southern Denmark / Biology; L. Weltje, University of Southern Denmark / Biology; L. Weltje, University of Southern Denmark / Biology; L. Weltje, University of Southern Denmark / Biology.

Interference of hepatotoxicity with endocrine activity in zebrafish (Danio rerio) discussed will include several experiments using the OECD fish embryo acute test (FET) and chronic exposures to juvenile and mature zebrafish. Interactions with the endocrine system of fish is crucial for understanding the risk associated with chemical exposure, as endocrine-related pathways are involved in growth, development and reproduction. These endocrine disrupting (ED) effects have been well characterised in aquatic vertebrates and mammals due to the well-defined endocrine system and substantial research effort in the last decades, but knowledge on ED effects in a larger range of species are still poorly characterized. Lack of knowledge on ED effects in invertebrates is currently a major limiting factor to properly perform risk assessment of endocrine disrupting chemicals (EDCs) across taxa. The present project has focussed on developing Adverse Outcome Pathways (AOPs) for EDCs in aquatic crustaceans, and applying these to assess the hazard and risk of ecologically relevant complex mixtures of pollutants. Although several ED mechanisms have been proposed to be of relevance for crustaceans, perturbations of endocrine processes related to chemical interactions with the ecdysone receptor (eGFr) and the Juvenile Hormone (methyl farnesoate) receptor (MRJ) have been identified to be relevant. The present paper focuses on the application of AOPs to (1) develop linkage between endocrine mechanisms and adverse outcomes, (2) identify knowledge gaps and inform testing strategies, (3) identify sensitive species/taxa, (4) identify likely define toxicity endpoints suitable for Integrated Approaches for Testing and Assessment, IATA, 5) identify potential EDCs and 6) practical implementation of AOP in-based Risk assessment. The EDC-related relevant exposure scenarios. Acknowledgement - Funding from RCN-221455 A Adverse Outcome Pathways for Endocrine Disruption in Daphnia magna, a conceptual approach for mechanistically-based Risk assessment (www.niva.no/edrisk), RCN-268294 “Cumulative hazard and risk assessment of complex mixtures and multiple stressors (www.niva.no/mixrisk)” and EU-FP7 project SOLUTIONS (http://www.solutions-project.eu/project/).

661 Assessing impacts of place-based mixtures of emerging contaminants on endocrine activity and adverse outcome pathways: comparisons of different life stages R. Klaper, University of Wisconsin-Milwaukee / School of Freshwater Sciences; J. Crago, Texas Tech University/TIEHH / Environmental Toxicology The Institute of Environmental and Human Health TIEHH Emerging contaminants often appear as mixtures of differing concentrations across a landscape. Traditional toxicological assays as well as receptor binding assays that examine the impact of the individual chemicals in isolation do not adequately detect all EDC mixtures and do not describe the collective impact of mixtures as there can be cross-talk among molecular pathways. Using place-based mixture concentrations of emerging contaminants in combination with multiple molecular initiating events from adverse outcome pathways can help to identify potential hotspots of potential environmental impact that cross multiple mechanisms of action. This talk will discuss the use of transcriptomics to modify the OECD fish embryo acute test (FET) and chronic exposures to juvenile and adults fish are being used to examine EDC pathway related disruption. Examples discussed will include several experiments using exposure mixtures representing those measured in several locations in Lake Michigan.

662 Interference of hepatotoxicity with endocrine activity in zebrafish (Danio rerio) L.A. Baumann, University of Heidelberg / Aquatic Ecology and Toxicology; H. Holbech, University of Southern Denmark / Biology; W. Løjte, BASF SE / Crop Protection Ecotoxicology; H. Schmidt-Posthaus, University of Bern / Institute of Animal Pathology; H. Segner, University of Bern / Centre for Fish and Wildlife Health; T. Braunbeck, University of Heidelberg / Centre for Organismal Studies Vitellogenin (VTG), a well-established biomarker for the diagnosis of endocrine activity in fish, is used in the OECD test guidelines 230, 239 and 234. A reduction of VTG production (mainly in males) is often a marker for androgenic or anti-estrogenic activity, whereas an increase of VTG (mainly in males) is associated with estrogenic activity. However, the synthesis of VTG may not only be modified by typical endocrine-related pathways, but also through non-endocrine-mediated processes. In particular, hepatotoxicity, i.e. toxicant-induced impairment of liver structure and function, can influence VTG as a biomarker, since it is synthesized in the liver. Changes in VTG caused by non-endocrine hepatotoxicity in a screening assay would unnecessarily trigger very labor-, animal- and cost-intensive higher tier testing (e.g. a fish life cycle test). Therefore, an intimate understanding of the interplay between primary endocrine-related and non-endocrine-related pathways influencing VTG production is crucial for the avoidance of false diagnoses. The present study is driven by the hypothesis that hepatotoxicity may interfere with VTG synthesis in the liver / test fish. Thus, we investigated the effects of two well-known hepatotoxics, acetaminophen (APAP) and isoniazid (INZ), on zebrafish (Danio rerio) in a 21-day flow-through exposure test according to OECD guideline 230. Various hepatotoxicity- and endocrine system-related endpoints were recorded: - mRNA expression of different endocrine-related (vgl1, vtg1 and esr1) and hepatotoxicity-related marker genes (fabp10a, apoa, cyp2k19 and cyp2s10) in the liver. - Cyp3a65 in the liver. - hyaluronic acid (a biomarker for liver toxicity) levels in head/tail homogenates; - liver histology and ultrastructure; Both APAP and INZ had different effects on exposed fish. While APAP did not cause any histopathological alterations in the liver, INZ significantly induced hepatocyte degeneration. VTG levels in APAP-exposed females were elevated, while no effect was observed in INZ-exposed fish. Likewise, the effects in the liver differed between both compounds and indicate that both did interact with different endocrine- and hepatotoxicity-related pathways. The results from both studies will further be evaluated with respect to their potential for the development of an adverse outcome pathway (AOP) for interference of hepatotoxicity with the VTG response in fish.

663 Single Pulse Exposure of Different Life Stages of Zebrafish to the Selective Estrogen Receptor Modulator Tamoxifen Citrate S. Kroesen, Fraunhofer IME / Institute for Molecular Biology and Applied Ecology; C. Schaefers, Fraunhofer-Institut / Ecotoxicology, E. Bruns, Bayer AG Division Bayer CropScience; M. Klimmer, University of Heidelberg / Centre for Organismal Studies AOP for interference of hepatotoxicity with the VTG response in fish. The results from both studies will further be evaluated with respect to their potential for the development of an adverse outcome pathway (AOP) for interference of hepatotoxicity with the VTG response in fish. The results from both studies will further be evaluated with respect to their potential for the development of an adverse outcome pathway (AOP) for interference of hepatotoxicity with the VTG response in fish.
664 A Tiered Approach for Screening Chemicals for Biomagnification Potential in Humans
A. Sangion, University of Insbsuria / Department of Theoretical and Applied Sciences (DiSTA); J.A. Arnot, ARC Arnot Research & Consulting / Adjunct Professor, Department of Physical & Environmental Science/Department of Pharmacology and Toxicology; P. Gramatica, University of Insbsuria / Department of Theoretical and Applied Sciences (DiSTA)
Bioaccumulation is a process in which the chemical concentration in an organism exceeds the concentration in the respiratory medium, the diet or both and is an integral aspect of hazard and risk assessment. Strong correlations between partitioning properties such as the octanol-water partition coefficient (Kow) and the octanol-air partition coefficient (Koa) and the toxicity of water-respiring and air-breathing organisms have led to Kow and Koa being the primary screening criteria for bioaccumulation assessment. However, primary biotransformation rate constants (ka) and half-lives (HLA) are also critical determinants of bioaccumulation. Here we present a tiered approach for screening the bioaccumulation potential of organic chemicals in air-breathing organisms. The tiered approach progresses from screening-level conservative assumptions based on Kow and Koa only to more realistic assumptions for, internal distribution, chemical properties and biotransformation (Tiers from 1 to 4 respectively). Biomagnification Factor (BMF) derived from a typical human diet as calculated by the Risk Assessment Identification And Ranking (RAIDAR) model is the metric for assessing bioaccumulation potential in air-breathing organisms of approximately 13W. Nichols, U.S. EPA / ORD NHEERL Mid Continent Ecology Division; B. Wetmore, U.S. EPA / National Exposure Research Laboratory; J.A. Arnot, ARC Arnot Research & Consulting / Adjunct Professor, Department of Physical & Environmental Science/Department of Pharmacology and Toxicology

665 Critical Evaluation of a Human In Vitro Biotransformation Rate Database: Case Study of Seven Chemicals
K.L. Foster, ARC Arnot Research and Consulting Inc. / Adjunct Professor, Trent University, Applications of Modelling & Quantitative Methods (AMOD); A. Looky, ARC Arnot Research and Consulting Inc.; J.M. Armitage, University of Toronto - Scarborough / Physical and Environmental Sciences; M. Embry, ILSI, 1W. Nichols, U.S. EPA / ORD NHEERL Mid Continent Ecology Division; B. Wetmore, U.S. EPA / National Exposure Research Laboratory; J.A. Arnot, ARC Arnot Research & Consulting / Adjunct Professor, Department of Physical & Environmental Science/Department of Pharmacology and Toxicology

Despite the fundamental value of biotransformation rate information, relatively few measured in vivo data are available for humans compared to the thousands of chemicals requiring evaluation. Reliable models, laboratory measured in vitro biotransformation rate data, and in vitro-in vivo extrapolation (IVIVE) methods can be applied to address in vivo biotransformation rate data gaps and, coupled with data confidence assessment methods, uncertainty and data utility. We have developed a new database of >11,000 human in vitro biotransformation rate estimates (half-lives, clearance rates and rate constants) derived from liver microsomal, S9 homogenate, and hepatocyte-based assays for >8,500 organic chemicals from the literature and publicly available databases (i.e., ChEMBL). The database is comprised primarily of pharmaceuticals and pharmaceutical candidates from various experimental sources. The organic chemicals in the database represent a broad range of physical-chemical properties (Log Kow= 4 to 13, Log Koa=0.01 to 47, Log Kow/Koa = 0.1 to 12, Kow in vivo = 0.01 to 1200 ml/g), span about 8 orders of magnitude. We developed and applied novel data quality assessment methods based on proposed standardized testing guidance to address variability and uncertainty in the database. The data quality assessment methods included compiling physical-chemical property data (e.g., Kow, pKa, water solubility) for all of the chemicals and applying a mass balance in vitro model. The resulting data quality scores (e.g., high or low confidence) may help identify datasets that are most appropriate for QSAR development and for other potential applications (e.g., bioaccumulation screening, prioritization). The score results are further examined in a case study of seven chemicals and the utility of high and low confidence biotransformation rate data, its merits and limitations for various use contexts, are discussed and overall key findings of the critical review of existing human in vitro biotransformation rate data are summarized.

666 Sediment-associated cyclic volatile methylsiloxanes: Biotransformation in a freshwater oligochaete and an estuarine polychaete
H. Selck, Roskilde University / Dept Science and Environment; R. Windfeld, Roskilde University / ENSPAC
Chemical regulatory legislation of organic contaminants is generally based on an assessment of the chemical potential to persist (P) in the environment, bioaccumulate (B) in biota, and possess potential toxicity. Applying standardized exposure setups (i.e., water-only exposure) as historically has been employed in environmental risk assessment, may overestimate bioaccumulation of hydrophobic organic contaminants (HOCs) in sediment-dwelling organisms because: 1) HOCs often accumulate in sediments to concentrations greatly exceeding the concentration in the overlying water; and 2) a number of papers illustrate that sediment-associated HOCs are available for uptake in benthic organisms. Alternatively, benthic organisms may be able to metabolize organic contaminants (i.e., biotransform), thus reducing their body burden. However, available information on the biotransformation capacity of benthic organisms is very limited. We conducted a number of experiments examining uptake and biotransformation of sediment-associated cyclic volatile methylsiloxanes (i.e., D4 and D5) in two deposit-feeding worms, namely, the estuarine polychaete, Capitella teleta and the freshwater oligochaete, Tubifex tubifex. This presentation will provide examples of how biotransformation capacity varies among the two benthic deposit feeders, and how biotransformation may reduce body burden and facilitate the removal of sediment-associated siloxanes. Including these factors in a hazard or risk assessment are likely to impact PBT assignment and categorization, and exclusion of benthic organism behavior may add compound uncertainty to predictions of bioaccumulation and trophic transfer.

667 Toxicokinetics and biotransformation products of diuron and 3,4-DCA in the developing zebrafish embryo (Danio rerio)
E. Jarosz, UFZ Leipzig / Bioanalytical Ecotoxicology; M. Kraus, Helmholtz centre Environmental Research UFZ / Department Bioanalytical Ecotoxicology; E. Küster, Helmholtz Centre for Environmental Research, Dept.Bioanalytical Ecotoxicology / Department Bioanalytical Ecotoxicology; T. Luckenbach, Helmholtz Centre for Environmental Research UFZ / Department Bioanalytical Ecotoxicology

Diuron is a commonly used phenylamide herbicide which acts by interrupting the photosynthetic electron transport chain. Like other phenylamide herbicides diuron is bio-transformed to 3,4-dichloroaniline (3,4-DCA) which occurs in plants, the liver of vertebrates and in soil. Fish embryos do not possess the same metabolic potential as adult fish. It was not determined so far if different embryo stages differ regarding toxicokinetics and 3,4-DCA and 3,4-DCA-TFA biotransform due to parent compounds or metabolites. We addressed the following questions: What are rates of uptake and elimination of diuron and 3,4-DCA in different zebrafish embryo stages? Is diuron biotransformed by the embryo via which metabolic pathway? Does the embryo’s chorion form a barrier for diuron and 3,4-DCA mitigating the compounds’ toxicities? Information on these aspects is valuable for the understanding of the toxicity of phenylamide herbicides to fish embryos. For determining tissue concentrations after different times of exposure, freshly fertilized zebrafish eggs were exposed to the EC20_12h, i.e., for diuron 2.86 mg/L and for 3,4-DCA 1.41 mg/L, pools of 7 embryos were shocked-frozen at 13 time points from 1.5 to 120 hours post fertilization (hpf). The test compounds were extracted from the embryo tissue with MeOH/H2O and quantified using liquid chromatography coupled mass spectrometry (LC/MSMS). Depuration of test chemicals from the embryo tissue was examined in five different developmental stages of embryos that upon exposure to chemicals were transferred to clean medium and then sampled after 0.5, 1.5, 3, 6 and 24 hrs. The tissue concentrations for diuron reached Tamas around 48 hpf, Tmax for 3,4-DCA was between 8 and 24 hpf. Based on the data for internal concentrations upon different exposure times uptake and elimination rate constants (k, k), were determined. Both elimination rates and residue of initial concentration after 24 hrs. of depuration differed between embryo stages. The search for possible metabolites showed that 3,4-DCA was transformed into 3,4-dichloroacetanilide in the embryo and two products of N-demethylation of diuron were found. This confirms that both phase I and phase II metabolic pathways are active in the first hours of embryo development and pinpoint the biotransformation capability of the zebrafish embryo at this early stage.
assessments of single chemicals. (iii) Development of PBTK models for environmental risk assessment of multiple chemicals. Generic PBTK models for single compounds in rainbow trout (*Oncorhynchus mykiss*), fathead minnow (*Pimephales promelas*), stickleback (*Gasterosteus aculeatus*) and zebrafish (*Danio rerio*) have been developed. Physiological description and parameters proposed by Nichols et al. [1] were updated by an extensive literature search. New mathematical functions were proposed to integrate the factors influencing the toxicokinetics (water temperature, growth dilution, reproduction cycle, ...). Default values for compound-specific parameters were estimated using QSAR models based on hydrophobicity [2, 3]. An optional interaction term was added to the mixture PBTK models for metabolic interactions such as competitive inhibition. Two case studies were selected based on the availability of toxicokinetic (TK) and toxicodynamic (TD) data for single compounds and for mixtures. In the first case study, the interaction between melamine and cyanuric acid was studied and in a second case study, the interaction between chlorpyrifos and permethrin was modelled. The models developed enabled to model interactions that are observed between exposure concentrations and final effects. The QSAR estimations of certain compound-specific parameters can compensate for the lack of data in fish. Extrapolation from one species to another with the various models developed can also help bridge gaps. [1] Nichols et al. 1990. Toxicol Appl Pharmacol 106:433-447. [2] Bertelsen et al. 1998. Environ Toxicol Chem 17:1447-1455. [3] Nichols et al. 2006. Aquat Toxicol 78:74-90.

669 Application of Aqueous and Dietary In-Vivo Bioaccumulation Tests to Determine Biotransformation Rates, Elimination Rates and other Bioaccumulation Metrics

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Bioaccumulation assessment is quickly evolving to respond to the need to conduct bioaccumulation assessment faster, better, cheaper while reducing animal use. In this presentation, we present novel methods for conducting both aqueous and dietary bioaccumulation tests that provide more information on bioaccumulation than currently used methods while also reducing animal use, effort and costs. Stream-lined aqueous bioconcentration test designs and dietary bioaccumulation studies following OECD 305 test guidelines are shown and discussed. A key element of the novel methods is the use of reference chemicals. In addition to experimental methods, we also present computational methods for deriving biotransformation rate constants, elimination rate constants and other bioaccumulation metrics with their associated error from the results of bioaccumulation tests. This involves an Excel worksheet, referred to as ADME calculator, that is specifically developed to interpret the results from aqueous and dietary bioaccumulation tests in terms of Absorption, Distribution, Metabolism and Excretion (ADME) rates including somatic and intestinal biotransformation rate constants, elimination rate constants, BCF and other bioaccumulation metrics. We further demonstrate that the application of reference chemicals can help to develop a full mass balance of the internal distribution of test chemicals in fish in the test and in the field under environment-specific exposure conditions. The application of the test results for determining exposure pathways of the test chemical under field conditions is illustrated. We conclude that bioaccumulation tests can become more effective in developing bioaccumulation profiles of chemicals when including reference chemicals.
**Poster Abstracts**

**Advances in environmental risk assessment of oil spills and offshore oil & gas operations (P)**

MO001  
An in-situ amphibian metamorphosis assay to evaluate oil spill-related toxicity in receiving freshwater systems  
R. Krohn, University of Calgary / Dept. of Ecosystem & Public Health, Faculty of Veterinary Medicine; J. Muscatello, Lorax Environmental Services Ltd; J. Smits, University of Calgary / Ecosystem & Public Health Faculty of Veterinary Medicine  
Divided brine mussels (dillims) transported from the oil sands in northern Alberta, consists of a mixture of chemicals, such as aromatic hydrocarbons, metals and other compounds, which may pose risks to wildlife and human health, if spilled into the environment. There is a major knowledge gap regarding remediation of oil spills into freshwater environments. The relative efficacy of different remediation strategies for these spill emergencies are untested. We have established an in-situ amphibian assay to serve as an indicator of health and recovery in freshwater ecosystems, which can be applied to assess risk and remediation efficacy. In spring 2017, Wood frog tadpoles were placed in 5, partially submerged cages (50 animals/cage), which were tethered to the peat-organic shoreline of Lake 260 of the International Institute for Sustainable Development-Experimental Lakes Area (ISD-ELA), Ontario, Canada. Tadpoles were fed and monitored every other day and were euthanized when >50% reached their metamorphic climax (the day of forelimb emergence), to perform gross anatomical examinations, sample collection and relevant biochemical analyses. Major outcomes: 1. Time to metamorphosis (an established, sensitive biomarker) 2. Mortality rate 3. Morphometrics (total body mass, length and hepatic mass) Analyses: 1. Hepatic detoxification effort (ethoxyresorufin-O-deethylase (EROD) enzyme activity); 2. Thyroid hormones levels (serum biomarkers of thyroid toxicity); 3. Triglyceride levels (reflecting body condition & energy stores). 4. Tissue contaminant levels (metals, PAHs) Baseline data for Wood frog development in Lake #260 were acquired in 2017, and potential pitfalls and solutions for the metamorphosis assay were identified. This assay will be used in the 2018 field season with the experimentalshorefront dillim and remediation strategies placed in Lake #260 at the ISD-ELA. In keywords: oil spill, endocrine disruptors, metamorphosis, Wood frogs

MO002  
APPLICABILITY OF RISK BASED, TIERED ASSESSMENT OF PRODUCED WATER DISCHARGE IN NIGERIAN SHALLOW OFFSHORE ENVIRONMENT  
M.G. Smij, Shell International; O. Anako, SPDC Nigeria Ltd  
The pursuit of excellence in managing risks associated with produced water discharges has led to continuous innovation of internationally acceptable risks assessment tools for determining the risks associated with produced water discharges. This study utilized a structured hazard analysis approach for the assessment of potential risks from water discharges. At the centre of this structure is a comparative analysis of predicted environmental concentrations (PECs) of chemicals and effluents to predicted no effect concentrations (PNECs) of environmental receptors. However, the determination of the likelihood and severity of effects is complicated and based on an integrated evaluation of several Lines of Evidence (LoEs). This study utilized risk-based assessment tools from Shell’s tiered assessment framework for discharges. This framework was developed based on international good practice and includes screening tools that allow for a rapid assessment of discharge properties and associated risks like, SMPE-GC and Microtox. In addition higher tier tools were applied like two way GCxGC, PETROTOX modelling and several levels of plume dispersion assessment. The objective is to determine the applicability of risk-based practice to the specific shallow offshore discharge, with possible replication to other shallow offshore or near shore discharges in Nigeria. It also assessed if discharges would be acceptable from an international perspective and whether indeed the risks are tolerable and as low as reasonably practicable (ALARP). Results of the assessment indicate that oil in water levels at or below 25mg/L are a low concern related to the environmental risk of the hydrocarbons in the PW discharge. Phenols and BTX were identified as the highest risk contributors but Tier-2 modelling indicated that these substances quickly dissipate after discharge. PETROTOX modelling showed that the hydrocarbon fraction in the PW could not fully explain the observed PW toxicity. This led to a recommendation for qualification and registration of offshore chemicals and identified a need for the alignment considerations of future Biological Monitoring programs to international protocols such as OECD and ISO. Application of smart screening tools (Tier-1) for frequent PBT monitoring to address variability and for tuning and focusing the larger Tier-2 and 3 assessments.

MO003  
Assessment of the biological impact of using chemical dispersants to remediate oil spills in different environmental conditions using zebrafish embryos  
Oil spills are a global concern due to their capacity to affect wide areas of the ocean and the well-being of the subsequent restoration of the ecosystem. Early life stages of fish are especially sensitive to oil spills as they are unable to flee the area and lack mature detoxification systems to withstand the chemicals. They also represent a direct link to population consequences and resilience. The addition of chemical dispersants can facilitate the dissolution of some chemicals present in the oil and make them more bioavailable. In addition, some chemical dispersants have been proven to be toxic chemicals. The impact of crude oil on a specific ecosystem and its recovery potential are determined by the biotic and abiotic elements of the ecosystem such as species composition, temperature, oxygen level and salinity. At low temperatures the persistence of hydrocarbons in the environment increases. Based on the standard OECD test with zebrafish embryos, we have tested the toxicity of the chemical dispersant FINASOL OSR52 and of the water accommodated fraction of a naphtenic North Sea crude oil produced with dispersant (WAF_{OIL+D}) or without dispersant (WAF_{OIL}) at different conditions of temperature and salinity. For WAF produced in marine water, polydimethylsiloxane (PDMS) sheets were incubated in the WAF_{OIL}, WAF_{OIL+D} and then used as passive dossers. Exposure to the dispersant caused 100% of mortality at concentrations ≥50 µg/L. Increased prevalence of malformations were present. The dispersant produced in different conditions. Although no clear differences were observed in relation with the temperature production of WAF_{OIL+D} / WAF_{OIL}, in general greater survival of organisms were observed in the presence of the dispersant. WAF_{OIL+D} and WAF_{OIL}. Zebrafish embryos appeared as a good model to study the toxicity of WAF depending on the temperature and on the addition of chemical dispersants. Funded by the EU H2020-BG-2005-2 project GRACE (grant agreement #679266), Spanish MINECO (NACE project CTM2016-81130-R) and MECD (FP6 grant to A.E.), the Basque Government (consolidated research group IT810-13) and the University of the Basque Country (IT11/37).

MO004  
Behaviour and effects of a marine diesel oil in a semi-static exposure experiment using mussels (Mytilus spp.) from the Baltic Sea  
R. Heikkinen, Finns Environmental Institute/ SYKE / Oil Research Centre; A. Ahvo, Finns Environmental Institute / Marine Research Centre; H. Kankaanpää, A. Reunamo, K.K. Lehtonen, K.S. Jørgensen, Finnish Environmental Institute / Marine Research Centre  
Marine diesel oil is produced and transported in large volumes in the Gulf of Finland and also used extensively as fuel in marine traffic in the Baltic Sea area. The heavily intensifying marine traffic in the area increases the occurrence of oil spills, smaller spills and leads to higher risk of major oil spills, which would certainly have drastic consequences to the local ecosystem. Chemical composition, mainly the polycyclic aromatic hydrocarbons (PAHs), can be more variable between the different diesel fuels, affecting the toxicity of the diesel to exposed marine organisms. The aim of this study was to determine the changes in the concentration of PAHs, acetylcholinesterase and other common biomarkers in the mussels (Mytilus spp.) exposed to a common type of low-sulphur marine diesel oil produced by Neste Oil’s Poroovo refinery in Finland. The diesel oil was applied to mussel aquaria as a water accommodated fraction (WAF). The exposure set-up consisted two replicate aquaria in each treatment; control, WAF-high and WAF-low, each with 200 mussels in 20 liters of artificial seawater (10°C). Water and WAF treatments were renewed every two days. Changes in PAH concentrations in water were constantly quantified using a TriOs Envirofluor HC-500 fluorometer sensor. Another sensor was used to collect auxiliary data on temperature, turbidity and chl a concentration (mussels fed with algae). Biomarkers of oxidative stress, biotransformation, neurotoxicity and bioenergetics were measured from mussels at the beginning of exposure and after a one week recovery period in clean water. Water and mussel tissue samples were also taken to chemical analysis of PAHs. Based on the sensor fluorescence data the initial PAH concentrations were ca. 30ng/L in WAF-high and 15µg/L in WAF-low treatments. In a semi-static system with mussels the concentrations decreased during 24h after which the level remained stable until the next water exchange. During the recovery period OHs occurred in water after every water exchange, suggesting significant release of PAHs from mussels (both from shell surfaces and internal pools). Differences between the treatments were observed in various biomarkers measured. Combined fluorescence, chemical and biomarker data give important insights to the fate and toxic effects of marine diesel oil in the northern Baltic Sea environment.

MO005  
Biliary PAHs and enzymatic biomarkers in the teleost Eugesurus brasiliensis along four tropical estuaries in the Brazilian Northeast  
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Human originated contaminants can appear diluted in estuarine and marine waters or accumulate in sediments. Chemical analysis provides key data on toxicant levels but gives limited inputs on their potential biological effects. The combination of biological responses with chemical data is essential to improve the assessment of environmental pollution. In this context, the use of benthic species for the assessment of biological effects of marine pollution is crucial for marine environment monitoring. In the Bay of Biscay, the common flatfish Solea solea sp., is considered to be a9ed as sentinel species in pollution monitoring programmes. The present study uses juvenile Solea sp. (23.24 ± 1.22 cm standard length) exposed to contamination conditions to better understand toxicity processes involved based on biochemical biomarkers and histopathology. Solea senegalensis was exposed to three different experimental set ups: (a) contaminated sediments; waterborne metal (Cd) and (c) waterborne organic pollutant (Benz(a)pyrene). A battery of biochemical biomarkers was analysed in samples of liver and brain: Catalase, glutathione S-transferase, acetylcholinesterase and superoxide dismutase. Exposure to contaminated sediments led to reduction of catalase, glutathione S-transferase and superoxide dismutase activities and induction of acetylcholinesterase activities. Exposure to waterborne toxicants provoked a reduction of catalase and glutathione S-transferase activities. Biochemical biomarkers in sole were sensitive enough to differentiate degree of response after three days of exposure. Histopathological responses were detected after long-term exposures showing higher prevalence of liver alterations such as hyperaemia, melanomacrophage centres and necrosis. The present laboratory experiments helped characterizing the impact of pollution in sole at different levels of biological organization and different time scales. Work funded by Spanish MINECO (CTM2012-39113) and Basque Government through Consolidated Research Groups fellowship (IT810-B). University of the Basque Country– UPV/EHU (UFI 11/37) and Basque Government through Consolidated Research Groups fellowship (IT810–B).

MO008
Biomarkers and Gene Transcription Variability in Perch in Reference Sites Used for Biomonitoring Studies
L. Forín, N. Askar, University of Gothenburg / Department of Biological and Environmental Sciences; M. Töpel, University of Gothenburg / Department of Marine Sciences; T. Österlund, Chalmers University of Technology / Mathematical Sciences; J. Parkonen, J. Sturve, University of Gothenburg / Department of Biological and Environmental Sciences

Perch (Perca fluviatilis) has been used in biological effect monitoring of point sources in Sweden for many years, for example in studies of effects of industrial effluents. Since 1988, perch has also annually been included in a program for integrated coastal fish monitoring in three reference sites along the Swedish east coast, sites characterized by no or minor local anthropogenic influences. Long term status monitoring of the reference sites enabled to follow the natural variability of physiological and biochemical endpoints (i.e. biomarkers) as this defines the changes of relevance in polluted sites. Using a set of physiological and biochemical endpoints (i.e. biomarkers) clear time trends for “early warning” signs of impaired health are noted in the perch from these three reference sites possibly as a result of increased baseline pollution. The data sets also show relatively large variations between years. To further investigate these time trends and to identify potential additional temporal variation in biological parameters, global gene transcription studies using RNA sequencing was performed. Perch collected in 2010 and 2014 were selected as they showed variation in several biomarkers such as the activity of the detoxification enzyme CYP1A (EORD), plasma levels of vitellogenin, markers for oxidative stress, white blood cells count and gonad sizes. The RNA sequencing workflow identified approximately 4,000 transcripts that were differentially expressed in the five sexually mature female perch collected in 2010 compared to the five individuals from 2014. Also principal component analysis (PCA) using all sequenced transcripts identified large differences in gene transcription as perch collected during the separate time points were clearly divided into two groups. Gene Ontology enrichment analysis showed that the differentially expressed genes were involved in biological processes such as innate immune responses, response to toxic substance, response to hypoxia and cholesterol biosynthetic process. In conclusion, differences in immune system parameters and responses to exposure of toxic substances have now been verified on two different biological levels (mRNA and protein) in perch collected in 2010 as well as 2014. Additional biological processes having temporal variation have been identified compared to the previous measurements of biomarkers.

MO009
Cellular and tissue-level biomarkers in mussels (Mytilus edulis) sampled in two different study areas in the Northern Atlantic
P. P. Sainz, M. P. Fernández-DETAIG, J. M. Balcázar, UPV/EHU / Departamento de Zoología y Biología Celular Animal; U. Izagirre, University of the Basque Country UPV/EHU / CBET Research Group Department of Zoology and Animal Cell Biology Faculty of Science and Technology and Research Centre for Experimental Marine Biology and Biotechnology P/E; I. Zorita, FUNDACIÓN AZTI; M. Soto, University of the Basque Country / Zoology and Animal Cell Biology. Research Centre for Experimental Marine Biology and Biotechnology P/E. The present study used juvenile Solea sp. (23.24 ± 1.22 cm standard length) exposed to contamination conditions to better understand toxicity processes involved based on biochemical biomarkers and histopathology. Solea senegalensis was exposed to three different experimental set ups: (a) contaminated sediments; waterborne metal (Cd) and (c) waterborne organic pollutant (Benz(a)pyrene). A battery of biochemical biomarkers was analysed in samples of liver and brain: Catalase, glutathione S-transferase, acetylcholinesterase and superoxide dismutase. Exposure to contaminated sediments led to reduction of catalase, glutathione S-transferase and superoxide dismutase activities and induction of acetylcholinesterase activities. Exposure to waterborne toxicants provoked a reduction of catalase and glutathione S-transferase activities. Biochemical biomarkers in sole were sensitive enough to differentiate degree of response after three days of exposure. Histopathological responses were detected after long-term exposures showing higher prevalence of liver alterations such as hyperaemia, melanomacrophage centres and necrosis. The present laboratory experiments helped characterizing the impact of pollution in sole at different levels of biological organization and different time scales. Work funded by Spanish MINECO (CTM2012-39113) and Basque Government through Consolidated Research Groups fellowship (IT810-B). University of the Basque Country– UPV/EHU (UFI 11/37) and Basque Government through Consolidated Research Groups fellowship (IT810–B). University of the Basque Country– UPV/EHU (UFI 11/37) and Basque Government through Consolidated Research Groups fellowship (IT810–B).
Marine Biology and Biotechnology PIE/UPV/EHU
Biomarker approach has been widely used in mussel monitoring programs for several years. However, up to now it has not been commonly used in high latitude study areas. In order to establish reference values of cellular and tissue-level biomarkers in the Northern Atlantic Ocean, mussels of two sizes (small, 2-3 cm; large, 3.5-4.5 cm) from selected polluted (commercial harbor & ports, WWTP dumping area) and reference sites in Tromsø (69º 40´N) and Trondheim (63º 26´N) were sampled in autumn of 2016 and late summer 2017. Different tissue-level biomarkers including cell type composition (VvVAS) in digestive gland epithelium, structural changes of digestive alveoli (MLR/OMET), relative proportion of digestive and connective tissue (CTD) and histopathological alterations in the digestive gland were measured. In addition, lipofuscin and neutral lipid acid accumulation, general degeneration, multifocal necrotic lesions, paracrine and atrophic, higher weighted prevalence values than in the reference site were recorded in the two polluted sites from Trondheim. Differences between the two mussel sizes were recorded in parasite burden, large mussels exhibiting a higher level of parasitization than small mussels. Lipofuscin accumulation was higher and neutral lipid accumulation lower in the polluted sites than in reference sites in both study areas. Lysosomal and lipofuscin areas were a direct hint to the presence of some toxic oil spills as biomarkers respond similarly in both study areas indicating the suitability of the selected biomarkers in order to be applied in the Northern Atlantic Ocean.

Acknowledgements: Work funded by, EU GRACE Project (Grant Agreement Number 679266), Basque Government (IT810-13) and UPV/EHU (UIF 11/37).

M0010
Cytotoxicity of the WAF of naphthenic North Sea crude oil with and without dispersant in hemocytes of the marine mussel Mytilus galloprovincialis (L.)
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Oil pollution coming from accidental oil spills and from activities related to oil production and its disposal is a serious threat to marine ecosystems. Mussels are long-lived benthic filter feeding invertebrates classified under oil spills may vary depending on environmental parameters such as temperature. There is a need to develop efficient tools to assess the risks of oil pollution and of oil spill response strategies, as the use of dispersants. The aim of this work was to apply an in vitro approach using hemocytes of the marine mussel Mytilus galloprovincialis as a model to evaluate the toxicity of the water accommodated fraction (WAF) of naphthenic North Sea crude oil produced at different temperatures (10, 15 and 20°C) with and without the dispersant Finasol OSR 52. In order to evaluate the contribution of the dispersant on the toxicity of the WAF plus dispersant, the cytotoxicity of the dispersant alone was also tested. Primary cultures of hemocytes were exposed in glass covered microplates to different dilutions of WAF (0.25, 2.5, 25, 50 and 100%) with and without the dispersant and to the dispersant alone at the same concentrations present in the WAF dilutions of oil plus dispersant (1.25, 12.5, 125, 250 and 500 mg/L). After 24 h exposure, cytotoxicity (MTT test) and ROS production were measured. WAF was moderately cytotoxic to mussel hemocytes. WAF produced at different temperatures showed similar cytotoxicity to hemocytes. A slight but significant decrease in cell viability occurred at 25, 50 and 100% WAF (p < 0.05) regarding temperature response. Multicellular burdens and atrophy, higher weighted prevalence values than in the reference site were recorded in the two polluted sites from Trondheim. Differences between the two mussel sizes were recorded in parasite burden, large mussels exhibiting a higher level of parasitization than small mussels. Lipofuscin accumulation was higher and neutral lipid accumulation lower in the polluted sites than in reference sites in both study areas. Lysosomal and lipofuscin areas were a direct hint to the presence of some toxic oil spills as biomarkers respond similarly in both study areas indicating the suitability of the selected biomarkers in order to be applied in the Northern Atlantic Ocean.

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M0011
Determination of inorganic cations and amines in wastewater, surface water, and neutralizing amine solutions by IC coupled with a single quadrupole MS
Inorganic cation and amine determinations are important to assess salt build-up in marine neutralizing solutions, or to meet regulatory discharge compliance from petroleum and municipal treatment plants wastewater. Additionally, municipal water plants require cation determinations to monitor secondary water characteristics. In the petroleum industry, alkylammoniums (monoethanolammonium, diethanolamine, and methyldiethanolamine) are used routinely to prevent corrosion during transportation to the refinery or to remove sour gases during the refining process. In processing plants watercourse contaminants such as amines are used to characterize and determine the next refining steps needed for oil and gas products received from various oil, gas, and fracking wells and to meet wastewater discharge requirements. Ion chromatography coupled to a single quadrupole mass spectrometer (IC-MS) is an ideal and economical way to determine and confirm cations and amines. Here we demonstrate cation, alkyamine, and alkylamine determinations in amine neutralizing solutions, amine wastewater, municipal wastewater samples, drinking, and surface water samples by cation-exchange separation followed by suppressed conductivity and mass spectrometry detections in a serial configuration. Cations, alkylamines, and alkylamines were determined in full scan from m/z 18 to 250 and individual SAs as bare ions and when further sensitivity is needed, as their hydrates adducts. Unlike earlier IC-MS methods for cation determinations, the new single quadrupole MS MS approach uses internal standards, and it is ideal for trace analyses in samples with complex matrixes, derivatization. Limit of Detections were single digit or double digit µg/L for most analytes. The experiments showed that typically sodium, ammonia, and primary amines were the primary contaminants in the scrubbing amines.

M0012
Distribution and ecological risk assessment of palm stearin in coastal marine environments of Hong Kong after an accidental pollution in Pearl River Estuary, South China
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On 3 August 2017, two container vessels collided in the Pearl River Estuary, southwestern of Hong Kong, leading to release over 1,000 tonnes of palm stearin into adjuvant in palm stearin, to assess its ecological risk to local marine ecosystems. This study, therefore, aimed to examine its contamination levels in seawater, sediment and animal samples collected from seven locations along the south coast of Hong Kong; determine its toxicities to selected marine organisms including microalgae (Isochrysis galbana and Chaetoceros gracilis), the copepod (Tigriopus japonicus), the fish (Gadus morhua), the shrimp (Penaeus monodon), and derive interim water quality guidelines (WQG) of the palm stearin and thereby assess its ecological risks to local marine ecosystems. Samples of the palm stearin, surface seawater, sediment and three intertidal gastropods were collected twice (within seven days and four months after the incident) in six locations along the south coast of Hong Kong. Fatty acids in these samples were detected using gas chromatography-mass spectrometry (GC-M/Quad) methods. This study represents the first comprehensive investigation on the ecological risk of the palm stearin in the world, and the results will facilitate informed decision-making by the environmental authority.

M0013
Ecological impacts of larvicidal oil on the marine ecosystem: implications on its management
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Mosquitoes are some of the most influential insects to mankind in the world due to their ability to transmit diseases to humans, resulting in millions of deaths every year. Prevention of mosquito-borne diseases and elimination of mosquitoes is important to protect human health. Among different methods of eliminating mosquitoes, the use of larvicidal oil is the most common way being employed in Hong Kong. However, larvicidal oil will be eventually released into the marine
MO014 Effects of a coastal oil spill on marine invertebrates and their potential to recover
M.F. Lemos, S. Silva, Instituto Politécnico de Leiria / MIRE I.P.leiria
There has been an increasing public concern and focus on marine contamination issues mainly due to the arisings of emergent pollutants, posing a major threat to human and environmental health. Still, the contamination by polycyclic aromatic hydrocarbons (PAHs) remains one of the most ubiquitous sources of pollution in the marine environment, being reported to elicit toxic, carcinogenic and mutagenic effects on marine biota. Moreover, the assessment of these impacts in coastal invertebrates after a spill, the extent of these effects and energetic trade-offs, potential recovery, and even which species to use is still deemed for an effective environmental contamination assessment. After an accidental industrial oil spill at the rocky shore of Peniche, Portugal in the summer of 2018, the water was analyzed during the low-tide for PAHs one week later and regularly throughout six months in the spilled beach and in 7 other rocky beaches in the vicinity. Also, at all locations, Patella depressa and Gibbula umbilicalis organisms were collected, and several biomarkers specific to the both species, the neurotoxins (i.e., the neurotransmitter > 1) from exposure to the oil were determined using Monte Carlo simulation, indicating that the current risk was unacceptable high. Hence, monitoring and control on the use of larvicide oil as mosquito control pesticide would be urgently needed to mitigate its ecological risks.

MO015 Effects of oil exposure on visual function in early life stage fishes
J.T. Magnusson, University of North Texas / Biology; A.J. Klurisgara, The University of Texas at Austin / Marine Science Institute; E. Allmon, The University of Texas at Austin; A. Esbaugh, University of Texas Marine Science Institute / Department of Marine Science; R.M. Heuer, University of Miami / Marine Biology and Ecology; J.D. Stiegitz, M. Grosell, RSMAS University of Miami / Marine Biology and Ecology; A.P. Roberts, University of North Texas / Advanced Environmental Research Institute
The Deepwater Horizon oil spill released millions of barrels of oil into the Gulf of Mexico, coinciding with peak spawning periods of ecologically important fish species, such as the mahi-mahi (Coryphaena hippurus), red drum (Sciaenops ocellatus), and sheepshead minnow (Cyprinodon variegatus). Downregulation of genes important in eye development and function, as well as morphological abnormalities have resulted from polycyclic aromatic hydrocarbons (PAHs) present in the oil at concentrations less than 10 μg/L, impacting fish vision. Mahi-mahi, red drum, and sheepshead minnow embryos were exposed to weathered crude oil and assessed for visual function using the flicker-fusion principle to monitor an optomotor response, with subsequent histological analysis taken of each larave's retina. Oil-exposed larvae exhibited a reduced PAH-dependent optomotor response with a reduction in retinal layers and neuronal connections that play an important role in visual function and image processing. The present study provides evidence that weathered crude oil affects the visual system in developing larval fish, and relates oil-induced histological effects to behavioral endpoints. This research was made possible by a grant from The Gulf of Mexico Research Initiative. Grant No: SA-1520, Name: Relationship of Effects of Cardiac Outcomes in fish for Validation of Ecological Risk (RECOVER).

MO016 Effects of oil spill on coastal seaweed in the Arctic
S. Wegeberg, Aarhus University / Department of Bioscience; J. Fritt-Rasmussen, K. Gustavson, Aarhus University / Department of Bioscience - Arctic Environment In case of an acute oil spill response operation, decision making regarding the operational response strategy and prioritization of biology at risk must be resolved. For that a Net Environmental Benefit Analysis, NEBA, is often performed to achieve the optimal environmental benefit with respect to choice of oil spill combat methodology and biology at risk. To provide data for assessing oil spill impacts in the Arctic areas, the effects of oil smothering of the macroalgae Fucus distichus, which is a dominant species in the intertidal zone of the coasts in the Arctic, as well as its self-cleaning potential by wash in sea, were studied. Effects of four different oil types were tested, including crude oil types, bunker oil and marine diesel. The studied effects on oil types were specified based on the origin of crude oil and refinery process, and hence may have different effects due to their physical and chemical characteristics. Phototrophic activity was measured as proxy for effect on growth and the self-cleaning potential was tested by wash in sea oil for smothered tips of F. distichus over a period of 2 weeks. The removal of the oils from the seaweed surface was considered as relatively fast (T 1/2 = 3-4 days). Depending of oil type, the oil inhibited or stimulated phototrophic activity. Marine diesel inhibited photosynthetic activity, whereas the three other oil types stimulated the activity. Thus, in general, the results indicated 1) that oil smothering was relatively fast washed off in the sea water; 2) that, depending on the oil type, phototrophic activity were stimulated or inhibited; and 3) that the photosynthetic activity was still affected (stimulated or inhibited) even after 14 days, although oil on the sea surface in the same study was removed much earlier. This work was funded by the European Commission Horizon 2020 programme and the Government of Greenland.

MO017 Effects of water accommodated fractions of crude oil on the Baltic Sea blue mussel Mytilus trossulus at different salinities
A. Ahvo, Finnish Environment Institute / Marine Research Centre; R. Turja, Finnish Environment Institute, SYKE / Marine Research Centre; A. Reunamo, Finnish Environment Institute, SYKE / Marine Research Centre; J. Nuutinen, Finnish Environment Institute / Laboratory Centre; K.K. Lehtonen, K.S. Jorgensen, Finnish Environment Institute / Marine Research Centre; A.G. Torreiro, Universidade Federal de Pernambuco / Department of Zoology; S. Wegeberg, Aarhus University / Department of Bioscience - Arctic Environment
In the Baltic Sea accidental oil spills are mainly combated using mechanical collection. However, this method is insufficient in harsh weather conditions such as high waves or in the presence of ice. The use of dispersants is an alternative countermeasure method but in the Baltic Sea their use is restricted by HELCOM recommendations since the chemically dispersed oil may cause severe toxic effects on marine biota. In addition, the use of dispersants is often questioned as the impact of water conditions is not well studied. In the present study, impacts of a crude oil and the dispersant Finasol 51 on marine biota were investigated under cold conditions (5°C) at two salinities corresponding to the German (15.0) and southern Finnish coastal areas (5.6). Baltic Sea blue mussels (Mytilus trossulus) were exposed to the water accommodated fraction (WAF) and dispersed WAF (WAF-D) of naphthenic North Sea crude oil in a semi-static aquarium experiment. Concentration of WAF or WAF-D in the aquaria was 5%. The mussels were sampled after 0, 1, 7 and 21 days of exposure, and analyzed for accumulation of polycyclic aromatic hydrocarbons (PAHs), and biological effects including acetylenochelinasterase, glutathione-S-transferase, catalase and glutathione reductase activities, lipid peroxidation, and protein carbonylation. In addition, changes in Mytilus-associated bacterial communities extracted from the gills and digestive glands of the mussels were investigated by sequencing of 16S rDNA genes and quantitative PCR targeted to bacterial PAH-degradation genes. Water samples from the exposure aquaria were taken for oil and PAH analysis. Dispersant augmented the amount oil in the exposure water with 0.13 mg/gL in 5.6 WAF compared to 44 mg/gL in 5.6 WAF-D (GC-FID, petroleum hydrocarbons C10-C40). A significantly higher oil concentration was observed at the lower salinity WAF-D water with 44 mg/gL oil at 5.6 and 1.82 mg/gL at 15. The higher salinity and WAF-D elicited more oxidative stress and neurotoxic effects already after one day of exposure. Mytilus-associated bacterial communities also varied depending on salinity and the use of dispersant. The results indicate that during the application of dispersants salinity plays a key role in the effect and oil concentrations in water as well as biological effects observed in the exposed biota. This should be taken into careful consideration when designing oil spill mitigation procedures in the Baltic Sea.

MO018 Multiple biomarkers on the estuarine guppy Poecilia vivipara to monitor two Integrated tropical estuarine ecosystems
A.G. Torrejón-Melo, UPPE Universidade Federal de Pernambuco / Department of Zoology; J.S. Silva, UFPE Universidade Federal de Pernambuco / Zoology; E. Zanardi-Lamardo, Universidade Federal de Pernambuco / Department of Oceanography; P.S. Carvalho, UFPE - Universidade Federal de Pernambuco / Zoología
Integrated approaches using biological responses in multiple organization levels are essential for environmental monitoring of tropical estuarines with ecologically relevant tools. The guppy Poecilia vivipara, native species with a broad tropical distribution, was utilized in such an approach, using in situ field exposures in cages.
(IS) and resident individuals (RES) collected close to the mouth of two tropical estuaries, Bacia do Pina Estuarine System (BES), and Barra de Jangada Estuarine System (BJES), in the Brazilian northeastern coast. This study was based on the analysis of water concentrations and internal accumulation of bile metabolites of polycyclic aromatic hydrocarbons (PAHs) by fixed fluorescence (FF), as well as biochemical responses related to the biotransformation of contaminants ethoxysterisorphin (Fraction I), benzo[a]pyrene (Fraction II), and aromatic hydrocarbons and polycyclic aromatic hydrocarbons (Fraction III). Behavioral activities related to swimming speed and resistance were also evaluated. Individuals grown in the laboratory were used for in situ exposure and also as controls (CON) for IS and RES. Significant contamination by PAHs was evidenced from both estuarian systems, with higher phenanthrene and chrysene concentrations in the bile of resident fish at BES, which in turn partially justified the significant induction of EROD and GST in these individuals. Resident fish at BJES showed high EROD and GST induction that cannot be explained by PAHs contamination, and suggests the presence of other contaminants with mechanisms of action similar to dioxins, possibly from a paper industry. Elevation of GST activity was detected in three of the four sites assessed on both estuaries, and loss of swimming resistance was verified on individuals exposed at the same sites, indicating a correlation between GST and this behavioral effect relevant to survival of the species. Indications of acetylcholinesterase inhibitors were not detected, except at the BPES inner region. This study shows the potential and feasibility of using the guppy *P. vivipara* on the evaluation and monitoring of pollution in estuaries along the Brazilian coast.

**MO019**

**NEW METHOD TO DETERMINE BTEX IN SOIL SAMPLES BY HPLC-DAD**

L. Silva, Universidade Federal do ABC / PROGRAD - CLD; C. da Silva, E.C. Lima, UFABC / CCNH; D. Rosa, UFABC / CECS

Benzeno, tolueno, elixehxileno e xileeno, commonly referred as BTEX, are components of fossil fuels, base case toxicants, and have negative impacts on the environment and human health. At fuel stations whose storage tanks are leaking, these substances may in contact with the soil and even reach the groundwater. In order to detect the concentration of these compounds in contaminated soils, gas chromatography (GC) is the most commonly used technique. In the present work it is proposed the use of high performance liquid chromatography coupled to diode array detection (HPLC-DAD) for the determination of BTEX in soil samples. A methodology was developed using as mobile phase methanol and H2O acidified with 250μL of H3PO4 (70:30, v/v), Eclipse XDB C18 column (5μm x 4.6 x 250mm), flow of 1.5 mL min^{-1}, λ = 205 nm and T = 50 °C. The analysis was carried out using the Agilent 1220 HPLC system equipped with an automatic injector, a column oven, and a diode array detector. Data was processed using Agilent OpenLAB A.01.05 software. A calibration curve for BTEX standards was constructed in 7 concentration levels: 1 to 68 ppb for benzene, 1 to 80 ppb for toluene, 1 to 80 ethylbenzene and 1 to 85 for xylene. The curves were submitted to inter- and intra-assay repeatability analyses. Standard curves with adjustments above 0.991 relative standard deviations (% RSD) of less than 1.9% were obtained. Reproducibility tests were performed with two solutions obtained from the standard solution. In the samples containing analytes from the soil contaminated with gasoline, % RSD was obtained below 6.5% and recovery rate was 68% for benzene and 75% for toluene, 78% for ethylbenzene and 78% for xylene. The method of soil analysis via HPLC is therefore efficient and as an alternative to be highlighted for analyses of soils contaminated with gasoline. It is intended to validate the methodology using appropriate protocols and apply it in contaminated areas for the verification of BTEX levels in the next step.

**MO020**

**Petroleum pollution of alluvial sediments near Sava river, Serbia**

M. Illic, IChTM / Department of Chemistry; S. Bulatovic, Faculty of Chemistry, University of Belgrade; T. Srdic Knudsen, IChTM / Department for Chemistry; M. Jelic, Institute of Chemistry, Technology & Metallurgy / Department of Chemistry; S. Miletic, J. Avdalovic, Institute of Chemistry, Technology & Metallurgy, University of Belgrade / Department of Chemistry; G. Devic, Institute of Chemistry, Technology and Metallurgy, University of Belgrade

Heating plant “New Belgrade” is located on the left coast of the Sava River, about 1km from its estuary in the Danube, and represents a potential source of petroleum pollution of alluvial sediments near Sava river, Serbia. The sampled material was organized in the layer structure with low content of organic matter. Extraction of petroleum pollutants from soil samples were done using the Soxhlet apparatus with dichloromethane. After extraction, the dichloromethane extracts were then fractioned by column chromatography into fractions of: saturated hydrocarbons (Fraction I), aromatic hydrocarbons (Fraction II), and polycyclic aromatic hydrocarbons (Fraction III). For monitoring changes in the vertical migration of petroleum pollutants, and the relationship of this migration with the soil characteristics, the group composition was determined for each borehole of all microlocations, taking into account their lithological profiles. Results of our research showed that in all samples the most represented were polar components (Fraction III), while the saturated hydrocarbon were least represented (Fraction I). This trend is almost unchanged in samples from different microlocation at different depths. It can be concluded that composition of petroleum pollutants can be unchanged through the alluvial sediments up to 15m depth and they can reach the underground waters, consequently disturbing the quality of the underground waters. A comparison of petroleum pollutants from samples collected in the vicinity of a heating plant in New Belgrade (Serbia) – influence on the quality of the surrounding soil and sediments. 16th European Meeting on Environmental Chemistry, EMEC16, Book of Abstracts. November 30 – December 03. 2015. Torino, Italy. Acknowledgements This work was supported in part by Ministry of Education, Science and Technological Development of the Republic of Serbia, Project No: III 43004.

**MO021**

**Prey capture to male aggression: the role of ecologically relevant behaviours in the assessment of complex petroleum based contaminants.**

D. Philibert, University of Alberta / Biological Sciences; D. Lyons, C. Philibert, University of Alberta; K.B. Tierney, University of Alberta / Biological Sciences

Crude oil and its associated by-products are ubiquitous in the aquatic environment due to both natural and anthropogenic sources (i.e. oil seeps and rivers flowing over surfacite bitumen, and pipeline ruptures, grounded ships, storage tank leaks and drainage from industrial and agricultural activities). Exposure to oil-based contaminants did not impair offspring growth, but instead altered the variation in behavioral phenotypes present in the population of exposed fishes. Previous studies suggest cortisol can be associated with behavioural phenotypes, and that developmental cortisol levels may pre-determine the functional phenotype found in a population of exposed fishes. Complex behaviours are sensitive sub-lethal endpoints that could be used in the risk assessment of contaminant mixtures. The inclusion of complex behaviours in toxicological studies brings ecological relevance to a biomarker dominated field.

**MO022**

**Risk-Based Approach: Assessment of Offshore Discharge Waters**

K. Wadhia, National Oilwell Varco (NOV) / Environmental; O. Pelz, BP / Gulf Coast Restoration Organization; S. Cousins, BP

In 2012, OSPAR (Oslo and Paris Conventions) adopted the recommendation 2012/5 for a ‘Risk Based Approach (RBA)’ to the management of Produced Water (PW) discharges from offshore installations’. The application of the RBA recommendations (2012/5) is implemented by the UK’s regulator, the Department for Business, Energy and Industrial Strategy (BEIS). The objective of the RBA is to assess the environmental risk of a PW discharge in the OSPAR maritime area. This is achieved by analysing the effluent and added substances to obtain a measure of the environmental risk. Previous studies suggest cortisol can be associated with behavioural phenotypes, and that developmental cortisol levels may pre-determine the functional phenotype found in a population of exposed fishes. Complex behaviours are sensitive sub-lethal endpoints that could be used in the risk assessment of contaminant mixtures. The inclusion of complex behaviours in toxicological studies brings ecological relevance to a biomarker dominated field.

**MO023**

**Petroleum pollution of alluvial sediments near Sava river, Serbia**

M. Illic, IChTM / Department of Chemistry; S. Bulatovic, Faculty of Chemistry, University of Belgrade; T. Srdic Knudsen, IChTM / Department for Chemistry; M. Jelic, Institute of Chemistry, Technology & Metallurgy / Department of Chemistry; S. Miletic, J. Avdalovic, Institute of Chemistry, Technology & Metallurgy, University of Belgrade / Department of Chemistry; G. Devic, Institute of Chemistry, Technology and Metallurgy, University of Belgrade

Heating plant “New Belgrade” is located on the left coast of the Sava River, about 1km from its estuary in the Danube, and represents a potential source of petroleum pollution of alluvial sediments near Sava river, Serbia. The sampled material was organized in the layer structure with low content of organic matter. Extraction of petroleum pollutants from soil samples were done using the Soxhlet apparatus with dichloromethane. After extraction, the dichloromethane extracts were then fractioned by column chromatography into fractions of: saturated hydrocarbons (Fraction I), aromatic hydrocarbons (Fraction II), and polycyclic aromatic hydrocarbons (Fraction III). For monitoring changes in the vertical migration of petroleum pollutants, and the relationship of this migration with the soil characteristics, the group composition was determined for each borehole of all microlocations, taking into account their lithological profiles. Results of our research showed that in all samples the most represented were polar components (Fraction III), while the saturated hydrocarbon were least represented (Fraction I). This trend is almost unchanged in samples from different microlocation at different depths. It can be concluded that composition of petroleum pollutants can be unchanged through the alluvial sediments up to 15m depth and they can reach the underground waters, consequently disturbing the quality of the underground waters. A comparison of petroleum pollutants from samples collected in the vicinity of a heating plant in New Belgrade (Serbia) – influence on the quality of the surrounding soil and sediments. 16th European Meeting on Environmental Chemistry, EMEC16, Book of Abstracts. November 30 – December 03. 2015. Torino, Italy. Acknowledgements This work was supported in part by Ministry of Education, Science and Technological Development of the Republic of Serbia, Project No: III 43004.
MO023
Risk-based assessment of produced water discharges - need for alignment
M.G. Singh, Shell International
Produced formation water is the main waste stream from upstream oil and gas activities. For offshore installations, next to produced water reinjection (PWRI), discharge of treated produced water is a commonly used disposal route applied in line with ALARP principles (As Low As Reasonably Practicable). In order to properly manage produced water discharges, a variety of principles have been adopted in national and international regulatory frameworks focusing on e.g. the oil in water content, toxicity of produced water, PBT characteristics of applied offshore chemicals, environmental monitoring, etc. The onus is on the industry to comply with regulations in the country of operation, to properly manage the risk of produced water discharges and to relieve any concerns over the potential environmental effects in the receiving marine environment. For this purpose the industry is applying a diversity of tools and methods within the framework of risk-based assessment (RBA). Tools and methods range from simple (tier 1) screening tools to comprehensive (tier 3) field verification programs and include among others; chemical analysis, determination of PBT characteristics through whole effluent toxicity studies and modelling, dilution screening and 3D dispersion modelling, etc. Within those practices a wide range of risk endpoints are being applied, each with their own level of conservatism. Without harmonization of endpoints it is difficult to interpret when risks can considered to be adequately controlled. For example, different approaches base the assessment on either chronic or acute toxicity data. And where one method considers an acceptable mixing zone of 500m, the other might apply 100m (USEPA). This hampers straightforward comparison of results of risk-based assessments across industry and regulatory frameworks. Different objectives of the assessment undertaken might, however, be a reason for existing differences in methods. It is, therefore, crucial that for each assessment method a corresponding assessment objective is formulated and communicated. This presentation will provide an overview of RBA practices currently applied by the industry to offshore produced water disposal, showing main assumptions, input requirements, risk endpoints applied and corresponding assessment objectives. Examples will be used to highlight the need for further harmonization of approaches. Development of industry guidance including a common tiered framework for RBA is suggested as a first step to achieve this.

MO024
Spatial and temporal analysis of the risks posed by total petroleum hydrocarbon and trace element contaminants in coastal waters of Kuwait
E. E. Nicolau, Cefas Lowestoft Laboratory / Environment and Ecosystems
Nine trace elements including As, Cd, Cu, Fe, Hg, Ni, Pb, V and Zn, and total petroleum hydrocarbon were analysed in whole effluent obtained from off-shore oil platforms and stations since 1984 from Kuwaiti coastal waters. Here it was investigated whether concentrations of these determinants are at levels above Kuwaiti and internationally established assessment criteria (AC). The results indicate that Cu and Cd had the most Kuwaiti AC breaches over time. Comparing the data of the last sampled year to the least stringent international AC, then Cu and Cd showed breaches at all stations. The trend in trace metal contamination is highly significant for Cd and Hg. No determinant measured showed a significant upward trend, indicating that water pollution for these contaminants is not a worsening situation. However, further sampling should be carried out to confirm these findings, especially at shoreline locations, where routine monitoring ceased in 2011 to investigate any recent changes.

MO025
Temperature-dependant toxicity of Naphthenic North Sea crude oil WAF, dispersant and their mixture: sea urchin bioassays
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Maritime traffic and oil platforms in the North and Baltic Sea have been growing due to the year-round drilling of oil and gas. Risk of spill, due to the harsh conditions at sea, and driven changes in water temperature could modify the potential toxicity of spill products including chemical dispersants. The impact of crude oil water accommodated fraction (WAF) and dispersants have been widely studied but their potential toxicity in accordance to the elctron micrograph of Lamark (1758) were used. In this study, larvae abnormalities and reduced larval growth indicate that toxicity was lower in crude oil WAF than in dispersed WAF, with their mixture in between for all the temperatures tested. Acknowledgement – This work has been funded by the EU H2020-BG-2005-2 project GRACE (grant agreement number 679266), Spanish Ministry of Education, Culture and Sport (PhD fellowship LDM FPU15/05317 grant) and the Basque Government (Consolidated Research Group GIC10T13).

MO026
Temporal variability of acute toxicity of Produced Formation Water discharged from offshore platforms: the responses of sea bass (Dicentrarchus labrax L., 1758) larvae
L. Mariani, CEFAS Lowestoft Laboratory / Environment and Ecosystems; A. Magaletti, B. Di Lorenzo, F. Onorati, C. Virno, Lamberti, ISPRA Institute for Environmental Protection and Research The Higher Institute for Environmental Protection and Research (ISPRA) is responsible for the evaluation of the potential environmental impact on marine ecosystem caused by the Produced Formation Water (PFW) discharged from Italian gas offshore platforms. A multidisciplinary approach has been applied through the monitoring of chemical-physical characteristics of water and sediment, matched with biological investigations, such as ecotoxicological bioassays on bacteria, algae, rotifers, crustaceans, echinoderms and fishes. The PFW is an effluent containing complex mixtures of contaminants, the composition of which may change with time. It is therefore necessary to analyse a large number of samples taken over a long period of time in order to adequately assess the toxicity of this effluent. The present paper addresses a specific toxic within the whole study; the variability of the acute toxicity responses of fish to PFW collected on two offshore gas platforms in three years (2003-2005). More sensitive life stages (post larvae of 25-45 days old) of European sea bass (Dicentrarchus labrax L., 1758) were used. Tests were performed over 24h and 96h and the dilutions, 6.25-12.5-25-50-100, containing PFW as it is highlighted by standard deviation values of LC50 data: exposure 24h (25.61 ± 7.02% PFW); 96h (10.84 ± 3.37 % PFW). In accordance with GESAMP recommendations (2007), the work stresses the importance of accurate estimates and measures of oil inputs into the sea, by increasing the number and frequency of samples needed to estimate the environmental hazard of PFW. The experience with the use of biomarkers as Risk Indicators in Environmental Risk Assessment of oil based discharges offshore
S. Sampi, International Research Institute of Stavanger / Environment; E. Lyng, D.M. Pampam, International Research Institute of Stavanger / Environment
Department

An approach to integrate biomarkers into probabilistic risk assessment has recently been developed and published regarding oil based discharges offshore. The main purpose has been to enable the use of monitored biomarker responses offshore as Risk Indicators in the procedures for Environmental Risk Assessment of produced water (PW) discharges. The principles of the approach and experiences obtained in applying it to existing oil field monitoring data will be presented. The approach was tested in two projects, the Danish Effluent Monitoring Initiative and Norwegian Continental Shelf Water Column Monitoring (WCM) program on the Norwegian Continental Shelf for assessment of PW effects. Cases including both a typical PW discharge and an alternative discharge make the data set interesting for testing the interpretation capability of the approach. At the site with no discharge of PW at the time of the study, a significant contaminant load from historical sources was identified. The experiences gained are discussed in relation to contaminant sources, use of the approach to provide assessment criteria for biomarkers, and for the performance of the biomarkers as risk indicators in relation to assessed environmental risk.

MO029 Tissue-level biomarkers and histopathological alterations in mussels (Mytilus trossulus) from the Baltic Sea exposed to water accommodated fractions of crude oil

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The brackish Baltic Sea is a fragile ecosystem potentially sensitive to oil spills. Chemical dispersants are an effective method to mitigate coastal impacts of produced oil spills; however, oil treated with dispersants may have unknown toxic effects on biota. The Baltic Sea blue mussel (Mytilus trossulus) is a particular variety of marine mussels adapted to low salinity. Early winter mussels were collected scuba diving in Tvärminne (Finland) in November 2016, taken to laboratory facilities and acclimatized at the experimental temperature of 5ºC to two different salinity regimes, the local 5.6 and the artificially increased 15.0 representing the southern Baltic Sea. Mussels were exposed to water accommodated fractions (WAF) and chemically dispersed WAF (dispersant Finasol OSR 51) mixtures (WAF-D) and sampled at 0, 1, 7 and 21 d. Tissue level biomarkers were investigated to determine the following biological responses: cell type composition (volume density of basophilic cells, \( V_{BAS} \)) of the digestive gland epithelium, structural changes of digestive alveoli (mean luminal radius/mean epithelial thickness, MLR/MET), mean epithelial thickness/mean diverticular radius (MET/MDR), connective/ diverticular ratio (C/D), diverticula ratio (C/T), glycogen content (Glyc), and histological alterations in digestive gland, gonad and gills. \( V_{BAS} \) increased significantly after 1 d in mussels exposed to WAF and WAF-D at the salinity of 15.0, and decreased afterwards. MLR/MET changed markedly with exposure time at 15.0 whereas MET/MDR showed no response. High C/T values in mussels observed at the salinity of 5.6 indicate a poorer condition of the digestive gland at low salinities than in mussels acclimatized at 15 psu. Pathological responses (atrophy, necrosis, diverticula ratio (CTD), gonadal development and other histopathological alterations) were measured at exposure and recovery intervals during the exposure and recovery period. One approach to the interpretation of the responses is to identify the degree of change and to identify the type of response. Interpretation of the responses includes the assessment of the degree of change and the type of response. The purpose has been to enable the use monitored biomarker responses offshore as Risk Indicators in the procedures for Environmental Risk Assessment of produced water (PW) discharges. The principles of the approach and experiences obtained in applying it to existing oil field monitoring data will be presented. The approach was tested in two projects, the Danish Effluent Monitoring Initiative and Norwegian Continental Shelf Water Column Monitoring (WCM) program on the Norwegian Continental Shelf for assessment of PW effects. Cases including both a typical PW discharge and an alternative discharge make the data set interesting for testing the interpretation capability of the approach. At the site with no discharge of PW at the time of the study, a significant contaminant load from historical sources was identified. The experiences gained are discussed in relation to contaminant sources, use of the approach to provide assessment criteria for biomarkers, and for the performance of the biomarkers as risk indicators in relation to assessed environmental risk.

MO030 Toxicity of produced water from offshore oil production in Norway and corresponding polar and apolar fractions

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Regulation of produced water (PW) discharges on the Norwegian continental shelf is still based on the assumption that PW toxicity may be efficiently assessed, with coverage of conventional oil quantification methods based on traditional GC is limited when it comes to polar compounds that originate from produced crude oils. Furthermore, the use of treatment or production chemicals might also contribute to the overall PW toxicity. As a result, there may be discrepancies between measured concentrations of organic compounds and the total PW components that contribute to the toxicity. This has been collected as "toxicity from four oil platforms on the Norwegian continental shelf. PWs were selected from oil fields of different operational ages, which produce oils exhibiting different physical and chemical properties. Samples were subjected to extraction with dichloromethane, followed by fractionation into apolar and polar fractions using solid phase extraction, recovering 80 % of the GC amenable material in these fractions. The total extracts and fractions were thoroughly characterized using GC−MS, GC×GC−MS, LC−Orbitrap-MS, and by direct infusion FT−ICR−MS. The total PW extract, as well as the apolar and polar fractions were subject to acute toxicity tests using nauplii of the marine copepod Acartia tonsa. LC50 values for the total PW extracts ranged between 0.05−0.98 mg L−1 (based on total GC amenable fraction analysis). For the apolar fraction, the toxicity was mainly attributed to the apolar fraction of the fourth PW, with an LC50 of 0.05 mg L−1. For the PWs where toxicity mostly related to the polar fraction, this fraction spanned from 16−55 % of the total PW (GC amenable fraction analysis). For the PW where toxicity mostly related to the apolar fraction this was 35 %. This study indicates that PWs exhibit a toxicity profile that is different from typical crude oil, with cover, compounds are currently poorly characterized. Polar fractions may contain compounds not amenable to GC, or that contribute to the GC−based fractionation of oil in water. This suggests that PW toxicity is not directly correlated with the GC quantifiable compounds that are used for regulating discharges today. Further studies should be pursued with a wider array of PWs from a range of sources to determine if alternative methods of characterization are needed for regulation of PW discharges.

MO032 Toxicokinetics of oil components in Arctic copepods

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To comprehend the implications of large oil spills in the Arctic marine environment, we need a better understanding of the toxicokinetics of oil in true Arctic organisms. The central nervous system (CNS) has a long evolutionary chain, as well as its life history strategies and Arctic adaptation, makes it a relevant target for understanding the toxicokinetics of oil in Arctic copepods. To achieve this goal, toxicokinetic studies were conducted on Copepod (larvae, adults) to determine the uptake, metabolism, and excretion of oil components. The aim of the study was to assess the toxicokinetic properties of oil components in Arctic copepods, as well as to evaluate the effects of oil exposure on the copepod's life history. The toxicokinetic properties of oil components were studied in Arctic copepods to assess the uptake, metabolism, and excretion of oil components. The aim of the study was to assess the toxicokinetic properties of oil components in Arctic copepods, as well as to evaluate the effects of oil exposure on the copepod's life history. The toxicokinetic properties of oil components were studied in Arctic copepods to assess the uptake, metabolism, and excretion of oil components. The aim of the study was to assess the toxicokinetic properties of oil components in Arctic copepods, as well as to evaluate the effects of oil exposure on the copepod's life history. The toxicokinetic properties of oil components were studied in Arctic copepods to assess the uptake, metabolism, and excretion of oil components. The aim of the study was to assess the toxicokinetic properties of oil components in Arctic copepods, as well as to evaluate the effects of oil exposure on the copepod's life history. The toxicokinetic properties of oil components were studied in Arctic copepods to assess the uptake, metabolism, and excretion of oil components. The aim of the study was to assess the toxicokinetic properties of oil components in Arctic copepods, as well as to evaluate the effects of oil exposure on the copepod's life history. The toxicokinetic properties of oil components were studied in Arctic copepods to assess the uptake, metabolism, and excretion of oil components. The aim of the study was to assess the toxicokinetic properties of oil components in Arctic copepods, as well as to evaluate the effects of oil exposure on the copepod's life history.
MO033 Two Dimensional Gas Chromatography for the analysis of polycyclic aromatic compounds and their alkylated homologues in environmental samples

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Polycyclic aromatic compounds (PACs) and their alkylated homologues are ubiquitous and known environmental contaminants. Due to their structural diversity and complexity of alkyl-substituted PACs, the resolution of individual alkyl congeners, especially those that are of environmental significance, is difficult if not impossible with conventional one-dimensional gas chromatography (GC). The peak capacity of the two-dimensional-GC has immensely improved analysis of these complex compounds in environmental matrices. In this study, the separation and detection of PACs were performed using a 30m cross-linked 5Sil MS (low polarity) and a 2m 17Sil MS (mid polarity) columns. A 2DGC-MS set-up was established for different sample matrices. This was achieved by varying the primary oven column length (30m to 60m) using the same column stationary phase (55μl MS (low polarity)) and a 2m 175μl MS (mid-polarity). To address a wider scope of PACs, samples were analyzed to include extracts of biota, used lubricating oil and coal samples. Resolution of individual isomers of interest was observed on the 30m primary column and much more evident on the 60m column. Undoubtedly, the peak capacity and vast database of information provided by 2DGC-HRTOF/MS for different sample matrices is an asset to the field of environmental research. This will aid isomer-specific measurements of known toxic alkyl homologues in environmental samples as well as proper identification of prominent ones. Also, PAHs and alkyl-PAHs profiling patterns have proven to be useful in source apportionment.

MO034 Using the hagfish (Myxine glutinosa) to study biological effects of a wreck filled with chemical munitions

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The sea bottom of the Skagerrak Strait (North Sea) contains ca. 45,000 tonnes of chemical warfare agents (CWA) dumped after the Second World War. Entire ships loaded with CWAs were intentionally sunk and are still laying on the deep bottoms (ca. 600 m) of the area in different states of deterioration by corrosion. The current status of the CWAs in the wrecks is unknown; if released into the environment they could negatively affect the marine ecosystem. In the present study, the leakage of CWAs and their possible biological effects on a marine organism, Myxine glutinosa, were evaluated. The species inhabits the studied depth range in the region. In this study, we used acetylcholinesterase activity as a biomarker to investigate the effects of CWAs. The study revealed that Myxine glutinosa from the wreck showed significantly higher acetylcholinesterase activity compared to the reference. This data indicated the presence of oxidized forms of CWA reductase and catalase activity) and for histopathological biomarkers, and muscle peroxidation, protein carbonylation, glutathione transferase, glutathione reductase and catalase activity) and for histopathological biomarkers, and muscle tissue was analyzed for acetyloaminotransferase activity. Chemical analyses were performed from muscle samples and separate whole fish samples, and the results indicated the presence of oxidized forms of CWA-related phenylacetic compounds in muscle samples. Established biomarker methods used widely in various fish species were shown here for the first time to be applicable also in hagfish. However, only minor differences in the measured biomarker responses between individuals collected from the wreck and the reference area could be observed. Based on this study, the hagfish is regarded as a suitable candidate for ecotoxicological studies of deep marine areas. More information on the biology of hagfish and the natural variability of their biomarkers is needed to distinguish true effects of hazardous substances.

Wildlife ecotoxicology: laboratory dosing studies to field

MO035 Seabird-derived contaminants and genotoxicity in Collembola from the Arctic

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Seabirds occupy high trophic positions and due to biomagnification their bodies contain high concentrations of many anthropogenic contaminants. In the Arctic they have been shown to function as important biovectors of contaminants from ocean to land. The tundra near bird cliffs is heavily influenced by nutritional and contaminant-enriched guano and is generally characterised by rich and diverse vegetation. In these areas, springtails (Collembola) contribute to a high proportion of bird biomass. Springtails play a vital role in the ecological function of the tundra such as decomposition and mineralization. The aim of this study was to determine the exposure, accumulation and effects of seabird-derived contaminants on Collembola. Two Collembola species and their habitat (soil/moss) were sampled at 7 sites with high, medium and low seabird influence in West Spitsbergen, Svalbard, and analysed for a wide range of organic contaminants, mercury (Hg), and stable isotope ratios of carbon and nitrogen (δ13C and δ15N, respectively). In addition, Collembola were analysed for genotoxic responses, i.e. the amount of DNA strand breaks and micronucleus frequency. Seabird influence (indicated by δ15N) and contaminant concentrations were indicated to be higher in soil/moss sampled closer to the bird cliffs (0–150 m) compared to further away (250–400 m) within the same site. When comparing among sites, however, no association between δ15N and contaminant load was found. The total contaminant loads in habitat samples were dominated by Hg, while no trend was found for organic contaminants. Contaminant concentrations in Collembola were dominated by Hg, followed by hexachlorobenzene (HCB), polychlorinated biphenyls (PCBs) or polybrominated diphenyl ethers (PBDEs), and chlorodane (CHLs). No association was observed between contaminant concentrations in Collembola and habitat. DNA fragmentation was higher in Collembola from sites with high seabird influence, compared to sites with medium and low. No differences in micromucular frequency (MN) was found between sites or species. This is the first study on MN in Collembola and should be further explored. DNA fragmentation, sensitivity to induced DNA fragmentation and micronuclear frequency were associated with both contaminant and contaminant load, increasing with concentrations of lower chlorinated PCBs and CHLs. The sensitivity to induced oxidative stress was negatively correlated with higher chlorinated PCBs (6-7 chlorine substitutions).

MO036 Higher contaminant and poorer condition in an Antarctic avian top predator from 2001 to 2013

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The Antarctic seabird, the south polar skua (Catharacta macormickii) has been considered low. However, recent investigations have shown that south polar skua (Catharacta macromickii) has the highest levels of biomagnifying contaminants among Antarctic seabirds. The present study quantifies OHC levels in south polar skua blood, and evaluate associations between contaminant occurrence, diet, trophic position, biological variables and day of sampling. Furthermore, the study investigates temporal change of organochlorine contaminant (OCs) by comparing previous data from the same colony, collected during the season of 2001/2002. South polar skuas were sampled during the breeding season of 2013/2014 in Svarthamaren, Dronning Maud Land, Antarctica. Whole blood was analysed for 87 OHCs of which 56 were detected. Stable isotope ratio of carbon (δ13C) and nitrogen (δ15N) in blood, were used to determine carbon source and relative trophic position, respectively. In 2013/2014, predominant contaminants were Mirex (8484 ng/g lw) and Hexachlorobenzene (HCB) (3561 ng/g lw). These levels were higher than those reported from other south polar colonies and Antarctic seabirds at similar ecological niches. Multivariate analysis indicated that skuas sampled late in the breeding season had higher concentration of perfluoralkyl substances (PFAS) and lower relative contribution of forage fish as prey, increasing with concentrations of lower polybrominated diphenyl ethers (PBDEs). Due to low intraspecific variation in δ13C and δ15N, no significant associations were found between OHCS and isotopes. However, lack of associations could also be due to influence of migration, wintering habitat and different turnover rates in OHCS and isotopes. Skuas from 2013/2014 had significantly higher concentrations of most OHCs and a lower body condition than skuas from 2001/2002. The study indicates that birds in 2013/2014 had a lower body condition than in 2001/2002.

Population assessments (P)

MO038
of south polar skua during the breeding season. Keywords: Antarctic, south polar skua, stable isotopes, temporal variation, OHC

MO037 Evaluation of malformations induced by a hospital effluent of Toluca (Estado de México) in Lithobates catesbeianus
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Hospital effluents are important from the ecotoxicological point of view and are an important source of pollutants emission, among which are pharmaceutical products, chemical residues, radioelements, disinfectants and heavy metals, among others, which are waste of daily activities and has reported that they can reach concentrations between 4 and 150 times higher than those detected in municipal effluents. In addition to this, they usually do not have adequate pre-treatment before their emission, so they can be mixed with other effluents from homes, industries and municipal wastewater, which can subsequently generate interactions, enhance effects and create synergies, which lead to induce adverse effects on the environment. So it is important to study. The Estado de Mexico is located in the center of the country and is considered the entity with the largest population and according to data reported in 2015, it has 1835 medical units of different levels of care. On the other hand, Lithobates catesbeianus is a species considered native of the state and has been proposed by the government as an alternative to food supplement (due to its protein content). Due to the aforementioned, the objective of this work was to evaluate the malformations generated by a hospital effluent of Toluca (Estado de México) in this species and compare with Xenopus laevis, a species that is used as a preferred biomonitor, using the frog embryo teratogenesis assay: Xenopus (FETAX). For this purpose, oocytes in mid-blastula transition were exposed for 96 h to six different concentrations of the effluent (0.1, 0.3, 0.5, 0.7, 0.9 and 1%); subsequently, the mean lethal concentration (LC50) effective concentration inducing 50% malformation (EC50), and the teratogenic index (TI) was obtained. Results indicates that lower concentrations of the hospital effluent induced slightly higher malformations and lethal effects in X. laevis (EC50=0.132%, LC50=0.508%; TI=3.8) and in L. catesbeianus (EC50=0.351%, LC50=1.431%; TI=4.0), the main alterations being microcephaly, cardiac and facial edema, malformations in the eye, notochord, tail, fin and intestine. However, the lightest concentrations of 0.1 and 0.3% significantly reduced the survival of this exposed to hospital effluent will be malformed in the absence of mortality compared to X. laevis. and therefore, can be considered as a sensitive and useful species to evaluate toxic effects of contaminants with the FETAX assay.

MO038 Monitoring fish health in a densely populated catchment in Central Germany
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In the frame of the joint project NiddaMan coordinated by the Goethe University Frankfurt/Main we investigated health parameters of fish from the River Nidda and its tributaries. For this purpose we used from different sources such as food, water and soil, therefore the toxicity of agrochemicals, as well as the patterns of use and consumption that is made used as indicators of water quality. Results showed LC50 values of 0.74 mg/l (0.63-0.89) (p= 0.001), exposing perinotryll to the standard medium. However, toxicity was almost inexistente when assays were done using natural river water. In this last case, the bioavailability of the fipronil was hypothesized to be reduced by natural substances present in the river water (solid suspended solids, organic matter, etc.). These results would contribute to a more realistic assessment of the environmental impacts of the use of this type of pesticides.

MO039 Multigenotypical toxicity of Fipronil to Folsomia candida
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Fipronil is a pesticide widely used on agricultural pest control, especially in sugarcane crops. This compound acts as an inhibitor of nerve signals in insects, and pose as a risk to non-target terrestrial organisms (i.e. the collombolan Folsomia candida), which plays important roles in the maintenance of soil quality. The main objective of this study was to evaluate the ecotoxicological effects on the reproduction of three generations of the Folsomia candida species when exposed to this compound over time, under a natural tropical soil. Test procedures were adapted from the ISO 11267 guideline. The chosen concentrations of fipronil were based on the recommended doses (RD) for the control of the pest Migelus flyrmus in sugarcane crops (RD = 1.3 mg of the commercial product / kg² of dw soil), what means 1.04 mg of fipronil / kg² dw soil. Concentrations tested were 0.06; 0.13; 0.26 and 0.52 mg fipronil kg⁻¹ of dry weight soil. The EC50 values were 0.21; 0.18 and 0.09 (±1) kg² / soil, for the first, second and third generation, respectively. According to the results, fipronil showed significant toxicity at low concentrations up to the third generation, causing effects on the reproduction and survival of Folsomia candida, and could be considered a highly dangerous pesticide for terrestrial arthropod organisms.

MO040 Fipronil effects on freshwater benthic algal communities
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Fipronil is a widely used broad-spectrum phenylpyrazole insecticide, effective against insects resistant to other agents as pyrethroids, organophosphates or carbamates. Accordingly, it has gained popularity worldwide as a pesticide in both agricultural and urban environments. Its wide use, resulted in the presence of fipronil in soil, surface and ground waters involving a risk to the environment and humans. In addition, humans and animals are also exposed to fipronil, by ingestion of products containing residues. Previous studies showed the toxicity of fipronil to aquatic freshwater organisms using model species, but there is scarce information about its impact on wild organisms. This study assesses, for the first time, the impact of fipronil on the photosynthesis of natural freshwater algal benthic community. This community – periphyton- is a key element of aquatic trophic chains, and is routinely used as indicators of water quality. Results show LC50 values of 0.74 mg/l (0.63-0.89) (p< 0.001), exposing perinotryll to the standard medium. However, toxicity was almost inexistente when assays were done using natural river water. In this last case, the bioavailability of the fipronil was hypothesized to be reduced by natural substances present in the river water (solid suspended solids, organic matter, etc.). These results would contribute to a more realistic assessment of the environmental impacts of the use of this type of pesticides.

MO041 Use of organophosphorus insecticides in agriculture lands, in a simple test
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Due to the human population increase and the consequent high demand for food, each day a larger area of the planet is dedicated to the practice of agriculture. Crops favor the reproduction of various organisms (invertebrates and vertebrates) that are combated with large amounts of pesticides, it are chemical compounds used extensively, and so all organisms are exposed from different sources such as food, water and soil, therefore the toxicity of agrochemicals, as well as the patterns of use and consumption that is made used as indicators of water quality. Results show LC50 values of 0.74 mg/l (0.63-0.89) (p< 0.001), exposing perinotryll to the standard medium. However, toxicity was almost inexistente when assays were done using natural river water. In this last case, the bioavailability of the fipronil was hypothesized to be reduced by natural substances present in the river water (solid suspended solids, organic matter, etc.). These results would contribute to a more realistic assessment of the environmental impacts of the use of this type of pesticides.
relationships (Pearson, R²=0.11) between the ChE and the weight or sex of the birds. The weight of the birds decreased on average 1.54 g after the exposure, possibly due to the stress of capture and the lack of adaptation to captivity. The house sparrow showed to be a species sensitive to the presence of OP compounds in the environment. It is possible that the house sparrow and other birds that co-inhabit the agricultural areas where malathion is applied, as well as other OP insecticides, are prone to develop different levels of intoxication and that in some cases their health condition is compromised.

**MO042** Implementation of a worst-case landscape scenario for population modelling of a fungicide applied in cereals

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In many cases EU member states prefer the use of country specific scenarios for the exposure or risk assessment of pesticides. This is sometimes requested due to specific geographical features or agricultural practice. In the present example, we demonstrate how the specific agricultural situation of the Netherlands was taken into account for the selection of landscape scenarios for use in population modelling. It is first shown how a country specific landscape scenario is developed. Then, the dose response obtained in a rat reproduction study with an azole fungicide is employed in a population-level risk assessment on small herbivorous mammals (Common vole, Microtus arvalis). The margins of safety obtained in that assessment would be expected under realistic worst-case field conditions.

**MO043** Biomonitoring and validation of non-invasive samples for the analysis of metals in freshwater turtles

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The mobilization of metals from the earth’s crust due to anthropogenic activities, such as mining, is one of the main contamination routes for wildlife. With the objective of biomonitoring and validating non-invasive methods for metal analysis in reptiles, levels of lead (Pb) and mercury (Hg) in blood, faeces and carapace scales of freshwater turtles (Mauremys leprosa, n=86) from historical mining areas (i.e. Valle de Alcudia-Sierra Madrona district for Pb, Almadén district for Hg) in Central Spain were analysed. The highest levels of blood Pb were found in the animals collected from the area of Valle de Alcudia-Sierra Madrona, more specifically from Solana del Pino, with an average (±SD) of 5.59±3.66 μg/g dry weight (dw). Individuals from this location showed the clearest evidences of oxidative stress, as estimated from the highest values of malondialdehyde (biodemer of lipid peroxidation) and superoxide dismutase activity. However, no correlations were found between these biomarkers and blood Pb levels. Similarly, the highest levels of Hg were detected in the animals collected from the area of Almadén, specifically from Almadenejos, with an average (±SD) of 8.83±8.44 μg/g dw. These individuals showed no evidences of oxidative stress, but presented increased activity of glutathione peroxidase and reduced glutathione levels relative to the rest of populations, which would indicate that antioxidant system is protecting from oxidative damage to occur. Blood levels of these two elements were above those reported as susceptible to cause sub-lethal effects in reptiles for the vast majority of terrains from the most contaminated sites (100% of terrains from Solana del Pino with blood Pb levels > 15 μg/dl; 70.3% from Almadenejos with blood Hg levels > 2.76 μg/g dw). Faeces and carapace scales obtained non-invasively correlated significantly with blood levels for the case of Pb (R ≥ 0.705, P < 0.001), but not for Hg (R ≤ 0.362, P ≥ 0.127). Thus, these samples could be used as non-invasive methods for the analysis of Pb bioavailability in M. leprosa, and by extension in reptiles, which will contribute to the development of ecotoxicology in reptiles, a group very little studied in this regard.

**MO044** An analysis of important life stages, exposure routes and test endpoints in amphibians and coverage by existing risk assessment regulatory requirements for plant protection products, part 1

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Concerns have been raised that the current risk assessment of plant protection products (PPP) may not sufficiently cover the risk to amphibians and reptiles. To address these concerns, the European Food Safety Authority (EFSA) has published a Scientific Opinion addressing the state of the science regarding the risk to amphibians and reptiles exposed to pesticides (EFSA 2017). A central task of the EFSA working group was to understand how well important life stages, exposure routes and endpoints are currently covered (or not) by the current risk assessment paradigm. We had four steps in this analysis: 1) Identify relevant life-stages, important exposure routes and possible endpoints with relevance to the specific protection goals (SPGs) defined for amphibians; 2) Evaluate the coverage by existing standard test guidelines with regard to these important stages/routes/effects; 3) Explore whether tests with surrogate species could provide suitable information allowing extrapolation to assess the hazard risk for amphibians; 4) Identify proposed non-standardized tests protocols suitable to address the gaps for specific protection goals defined for amphibians. Care was taken to address all relevant routes of exposure, life-stages, and short and long term effects. For amphibians, greater coverage or surrogacy exists for the aquatic larval stage and short term effects, with less coverage of the adult terrestrial stage, reproductive toxicity, and specifically for the potential exposure routes. To cover important life stages, exposure routes and effects, tests addressing dermal overspray and reproductive toxicity in amphibians may be needed. The analysis for reptiles will be presented in a separate poster. The concerns that the current risk assessment of pesticides may not sufficiently cover the risk to amphibians were supported after the analysis of the currently available data. The exercise provides a useful base for further research necessary to advance the ecotoxicological risk assessment of amphibians within the remit of the PPP authorization.

**MO045** European common frog (Rana temporaria) larvae show subcellular responses under field-relevant Bacillus thuringiensis var. israelensis (Bt) exposure levels used in mosquito control

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Bacillus thuringiensis var. israelensis (Bt) is presumed to be an environmental friendly agent for use in either health-related mosquito control or the reduction of nuisance associated with mosquitoes coming from temporary flooded wetlands. Amphibians co-occurring with mosquito larvae in these wetlands may be exposed to Bti products several times during their breeding season. Up until now, information regarding effects on the non-targeted group of amphibians has to be regarded rather inconsistent. On this account, we evaluated how repeated exposures to frequently used Bti formulations (VectoBac®2AS, VectoBac®WG) in field-relevant rates affect European common frog (Rana temporaria) larvae. In a laboratory approach, tadpoles experienced exposure conditions similar to realistic mosquito control in the Upper Rhine Valley (Germany). We assessed potential effects with regard to enzymatic biomarkers (glutathione-S-transferase, glutathione reductase, acetylcholinesterase), development, body condition and survival until the end of metamorphosis. Regardless of the formulation, delivery form or application rate, tadpole survival rates and time to metamorphosis were slightly reduced after repeated Bti exposures, while body condition was similar throughout the treatments. Furthermore, Bti induced significant increases of all enzymatic activities irrespectively of the applied field rate and formulation, indicating oxidative stress as well as unspecific neurotoxic effects. Repeatedly executed Bti applications, especially acting on early developmental stages, seem to increase the risk for adverse effects. The examination of several biochemical markers is needed to evaluate the ecotoxicological risk of Bti for amphibian populations, especially in the context of worldwide amphibian declines. Following the precautionary principle, the implementation of certain thresholds for application numbers and intervals should be considered in order to ensure environmentally friendly mosquito control programs, especially in areas originally designated for nature conservation.

**MO046** Influence of salinity and temperature on tadpoles of Xenopus laevis

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Worldwide coastal low-lying ecosystems are experiencing an increased salinity due to sea water intrusion. Coastal wetlands are of economic and environmental importance and areconsidered very vulnerable to salinity changes. In this context, the present study aimed at evaluating the influence of temperature on the adverse effects that increase of mean temperatures, until 2100, and consequent sea level rise, is foreseen an increase in the number of coastal ecosystems suffering from such salinization. Among coastal ecosystems that will be impacted with seawater intrusion are wetlands, which constitute hotspots of biodiversity and represent relevant ecosystems for amphibians. This class of ecosystems suffering from such salinization. Among coastal ecosystems that will be impacted with seawater intrusion are wetlands, which constitute hotspots of biodiversity and represent relevant ecosystems for amphibians. This class of ecosystems suffering from such salinization. Among coastal ecosystems that will be impacted with seawater intrusion are wetlands, which constitute hotspots of biodiversity and represent relevant ecosystems for amphibians.
with increasing salinity. Although significant interactions between temperature and salt concentration were registered, a pattern of influence in the temperature was not observed. Adverse effects were observed at the lowest tested salinity levels, which suggest that these species are highly vulnerable to small salinity increases and would be at high risk under seawater intrusion scenarios. Keywords: Salinity; toxicity; amphibia

MO047 EFFECTS OF THE EXPOSURE OF LARVAS OF Dendropsophus colombianus (ANURA: HYLIIDAE) TO WATER CONTAMINATED BY ANTROPOGENIC ACTIVITIES IN A RIVER BASIN OF THE COLOMBIAN ANDES

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The pollution generated by agriculture, livestock and mining have impacted the watersheds in the Colombian Andes. Amphibians have been used to evaluate this contamination due their biphasic lifecycle, which has made them ideal models in aquatic ecotoxicology. The objectives of this work were: 1) to determine if tadpoles of D. colombianus exposed to contaminants of agricultural, livestock and mining activities (with mercury: Hg, and with mercury and cyanide: Hg/CN) varied in the snout-vent length (SVL), tail length (TL), head width (HW), and body weight, and 2) to evaluate the effect of exposure on metamorphosis and behavior of the larvae. The AMPHTOX protocol was followed using ten larvae in each of the treatments and in the control, which were exposed from the moment of hatching to complete metamorphosis. Significant differences were found in the LRC between the larvae of the control and the Hg/CN mining treatment (Z= -28.92, p= 0.000) and between Hg/CN mining and agriculture treatments (Z= 25.325, p= 0.001) after 50 days of exposure. Differences in LC were found between the larvae of the control and the Hg/CN mining treatment (Z= -25.57, p= 0.001), and between Hg/CN mining and Hg mining treatments (Z= 21.525, p= 0.009) in the same time. The weight did not show differences. The majority of larvae of the control and the agriculture treatment showed similar development rates, reaching stage 46 between days 60 and 75 of exposure. While tadpoles exposed to Hg/CN mining did not complete the metamorphosis and reached stage 42 between days 50 and 55. Surface flotation was the activity that was most stable at the time of exposure, presenting percentages of individuals greater than 60% in the control (81%), and in the agriculture (70%) and Hg mining treatments (65%), between the last days of exposure evaluated: 22 to 28. It is shown that 1) the effects on growth of anuran larvae due to agricultural, livestock and mining contamination are similar. However, samples contaminated by mining produce smaller tadpoles than agriculture and livestock, and 2) tadpoles exposed to agriculture and livestock contamination, unlike other studies which registered times of metamorphosis, show a time of this process that approximates to the time that the species experimented in the control (134 days) and probably, to the time of this in situ.

MO048 Risks for amphibians and reptiles by dermal exposure to pesticides

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Amphibian and reptilian species are found in agricultural lands. Some inhabit water bodies either permanently or during some time of their life cycle. Others use predominantly the terrestrial environment. Some of these species migrate long distances through fields and some reside in fields and field margins where they can be exposed to pesticide residues in food items, water, soil and plant surfaces. A number of studies indicate that pesticide exposure can lead to severe impacts with up to 100% mortality from overspray at field rates for some pesticides. Dermal exposure was identified as an important exposure route in the EFSA opinion. The aim of the current study was to investigate different options to address the risk from dermal exposure by overspray and contact to soil and plant surfaces. Existing exposure models were reviewed with regard to their suitability for amphibian and reptilian risk assessment. This included comparison of parameters used in model calculations, comparison of sensitivity to dermal exposure and worst case exposure calculations to estimate risk to amphibian and reptile species. Highest dermal exposure from overspray and to compare it with exposure from contact to soil and plant surfaces. In addition a novel approach was developed to estimate the uptake from soil and plant surfaces. Mammalian dermal toxicity related to local effects and dermal adsorption data may be used as surrogates for reptiles but not for amphibians. The development of a test method investigating local effects on amphibian skin as well as the quantification of the inhibition of body burden following exposure via the dermal route is needed. Comparison of body burden by overspray to body burden by exposure to treated soil or foliage showed that the maximal body burden by overspray was lower than the maximal body burden by passive or active dermal exposure from soil or by contact to foliage. An approach was suggested which can combine oral and all dermal exposure routes in one overall body burden which could form the basis for a realistic risk assessment.

MO049 Evaluating the Role of Fish as Surrogates for Amphibians in Ecological Risk Assessment

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Ecological risk of chemical exposure to aquatic-phase amphibians is historically evaluated using surrogate toxicity data from standard fish species. Recently published meta-analyses indicate that amphibian ecotoxicology is not generally similar to that of fish, and amphibian and fish species are similarly sensitive to a range of chemicals. However, these analyses are limited because the amphibian data reported in the peer-reviewed literature are variable both with respect to experimental design and test species. In 2010, the U.S. Environmental Protection Agency began receiving ecotoxicity data for a standard amphibian test species (Xenopus laevis) as part of the Endocrine Disruptor Screening Program. Analysis of these data has informed to determine a determination of potential thyroid interaction within the context of other endocrine screening studies, they also contain valuable data on survival and growth that can be compared to existing fish data for a given chemical. We used this dataset to compare no observed adverse effect concentration (NOAEC) values for survival, body weight, and length data between fish and amphibians for 45 different pesticide active ingredients. Overall, the results indicate that fish are a reasonably good predictor of amphibian toxicity as there were no statistically significant differences in NOAEC values between the two groups for the endpoints examined. However, toxicity endpoints were lower in amphibians as compared to fish approximately half the time across chemicals, challenging the notion that fish are consistently more sensitive than amphibians. Disclaimer: Disclaimers: The views expressed in this report do not necessarily represent the views of the U.S. EPA or the United States.

MO050 Long-term survival of mancozeb exposed common vole populations from one to the following reproductive season

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Rodent field effect studies relevant for pesticide risk assessment typically take place during crop development from spring to autumn and thus within a single reproductive season. However, animal survival covering multiple reproductive seasons has only rarely been considered. This includes also microtine rodents such as the common vole (Microtus arvalis), as they are considered as rather short-lived vertebrates. However, overwintering individuals are important for the survival of local populations, and potential pesticide exposure during maturation might impact survival during seasonal bottlenecks in winter from one to the next reproductive season. Furthermore, from a regulatory perspective and due to the one-reproductive-season-duration of most field effect studies in wildlife, there is an increasing concern on potential long-term effects from exposure occurring delayed or that the reproduction in the following season might be affected by exposure during a previous application season. Against this background we monitored individually marked common vole populations from a long term effect study on spray applications of Dithane M-45 (Mancozeb 80% WP) during one reproductive season further on into the following reproductive season. The test item Dithane M-45 was applied four times in June according to Good Agricultural Practice at an application rate of 2 kg a.s./ha. Trapping and marking of voles in the same investigation plots was conducted until September, followed by further trapping until spring of the following year and the onset of the new reproductive cycle. Reproductive parameters recorded as indicators of potential long-term effects resulted in very similar patterns in treatment and control plots, and the data show no indication that common voles were negatively affected by multiple application of the test item also in the following year. Furthermore and up to now not documented for wild living common voles, the data gives evidence that free-living common vole individuals can reproduce for more than one reproductive season.

MO051 An analysis of important life stages, exposure routes and test endpoints in reptiles with regard to coverage by existing risk assessment regulatory requirements for pesticides

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Concerns have been raised that the current risk assessment of pesticides may not sufficiently cover the risk to amphibians and reptiles. To address these concerns, the European Food Safety Authority (EFSA) has published a Scientific Opinion on potential long-term effects from exposure occurring delayed or that the reproduction in the following season might be affected by exposure during a previous application season. Against this background we monitored individually marked common vole populations from a long term effect study on spray applications of Dithane M-45 (Mancozeb 80% WP) during one reproductive season further on into the following reproductive season. The test item Dithane M-45 was applied four times in June according to Good Agricultural Practice at an application rate of 2 kg a.s./ha. Trapping and marking of voles in the same investigation plots was conducted until September, followed by further trapping until spring of the following year and the onset of the new reproductive cycle. Reproductive parameters recorded as indicators of potential long-term effects resulted in very similar patterns in treatment and control plots, and the data show no indication that common voles were negatively affected by multiple application of the test item also in the following year. Furthermore and up to now not documented for wild living common voles, the data gives evidence that free-living common vole individuals can reproduce for more than one reproductive season.
non-standardized test protocols suitable to address the gaps for specific protection goals defined for amphibians and reptiles. Care was taken to address all relevant routes of exposure, life-stages, and short and long term effects. Reptiles have very little data with which to assess the coverage of important life stages and exposure routes. Current knowledge suggests that surrogacy based on bird or mammal data may not be appropriate for the juvenile and adult stages of reptiles, though data are very limited. Reptile eggs are not covered by any aspect of the current risk assessment paradigm, but it is unknown to what extent reptile eggs lay eggs in crop fields, and how likely exposures are to occur to eggs under realistic scenarios. Therefore, more data are needed to determine if this is a vulnerable life stage that needs specific consideration. The concerns that the current risk assessment of pesticides may not sufficiently cover the risk to reptiles were supported after the analysis of the currently available data. The exercise provides a useful base for further research necessary to advance the ecotoxicological risk assessment of reptiles within the remit of the pesticide authorization.

MO052
AmphิMove: Moving patterns and microhabitat selection of European anurans in agricultural landscapes
The current decline of amphibian populations on global and local scales is discussed extensively throughout the world. Studies suggest that enhanced application of plant protection products (PPP) is one of the main reasons that today amphibians represent the most endangered vertebrate group in Europe. The first version of a scientific opinion currently released by EFSA (European Food Safety Authority) highlights the need for including amphibians in the risk assessment of plant protection products. Specifics in terms of physiology (e.g., permeable skin) and ecology (e.g., aquatic and terrestrial life-stages in one single species) (life patterns and habitat preferences) make this taxon in particular vulnerable to pesticide applications in agricultural landscapes. It is emphasized that detailed ecological data of especially terrestrial amphibians is still under-represented but required for a comprehensive risk evaluation of all amphibian life-stages. The aim of the project AmphιMove is to fill the gap on terrestrial life-stage events of European anurans with focus on moving patterns and microhabitat selection in agricultural landscapes. At two study sites individuals of common toads (Bufo bufo) and common frogs (Rana temporaria) were caught at and around their breeding ponds, fit with a transmitter and afterwards tracked via radio-telemetry. Locations, biotic and abiotic parameters of the selected microhabitats were recorded daily. We show preliminary results of the first period of data collection for common toads from March to October 2017.

MO053
A quantitative AOP for activation of the aryl hydrocarbon receptor leading to early life stage mortality in amphibians and reptiles
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In order to advance the ecotoxicological risk assessment of aquatic and terrestrial AMPs, the reactivation assay to in vivo embryo sensitivities for an amphibian, the African clawed frog (Xenopus laevis) and a reptile, the common snapping turtle (Chelydra serpentina). Embryo-mortality was assessed in African clawed frog embryos exposed to serial concentrations of one of two DLCs: 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) or 2,3,4,7,8-pentachlorodibenzo-p-dioxin (PCDD). Embryo-mortality was assessed in common snapping turtle embryos exposed to serial concentrations of one of four DLCs: TCDF, PeCDF, 2,3,3’,4,4’,5-pentachlorobiphenyl (PCB 126), and 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD). Further, in vitro AHR transactivation assays were used to determine sensitivity to activation of the AHR1 isoform of African clawed frog and common snapping turtle to these selected DLCs. It is anticipated that this research will result in a single qAOP linking in vitro activation of the AHR to embryo-mortality with taxonomic applicability across phylogenetically diverse oviparous vertebrates, including birds, reptiles, amphibians, and fishes. This qAOP could guide more objective ecological risk assessments of DLCs to diverse taxa which are not easily studied, such as native species of reptiles and amphibians.

MO054
Do historically metal-exposed amphibian populations acquire resistance to lethal levels?
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Historically, exposure to heavy metals may have negatively impacted amphibian populations. However, little is known about whether the sensitivity to activation of AHR is adversely affected by historical exposure to heavy metals. For this reason, we aim to assess if metal-exposed populations are able to acquire resistance to lethal levels of heavy metals. For this, we are assessing resistance to 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) in the Japanese medaka (Oryzias latipes), a model organism for aquatic toxicology studies. We are using two strains of this species: one that was exposed to TCDD for several generations and another one that was not exposed. We are measuring the effects of TCDD exposure on different endpoints, such as survival, growth, and behavior. Our results will be compared to those of non-exposed populations to assess if resistance to lethal levels has been acquired.

MO055
Assessment of metal contamination levels and stress responses of endangered sea turtles of São Tomé and Principe
In São Tomé and Principe, sea turtles are an important component of the marine biodiversity. However, they are affected by various anthropogenic pressures, such as pollution, habitat loss, and overharvesting. One of the most significant threats is ocean pollution, which can lead to severe stress responses and health status in these animals. We aim to assess the level of metal contamination in the tissues of sea turtles of São Tomé and Principe and to determine their stress responses. For this, we are collecting samples of blood, liver, and kidney from wild sea turtles. We are measuring metal concentrations in these tissues and assessing oxidative stress biomarkers, such as lipid peroxidation and antioxidants. Our results will be compared to the levels observed in non-exposed populations to assess if these turtles are able to cope with metal pollution. This information will be crucial for the conservation management of these endangered species.
MO056 Ecotoxicology of Africa's three largest reptiles: POPS, metals, eggs, and eggshells
H. Biromann, North-West University / Unit for Environmental Science and Management; R. Nel, Nelson Mandela Metropolitan University / Department of Zoology; H. Kylin, Linköping University / Department of Thematic Studies Environmental Change; D. Govender, SANParks; M. Du Preez, North West University / Zoology

The Nile Crocodile (Crocodylus niloticus), Loggerhead Turtle (Caretta caretta) and Leatherback sea Turtle (Dermochelys coriacea) are the largest reptiles in Africa. The bioaccumulation and effects of metals and metalloids on large-bodied reptiles are less well known compared with birds and mammals, especially those from Sub-Saharan Africa. Globally, reptiles are experiencing declines, and pollution is one of the hypothesized reasons for the decline. The Nile Crocodile and Loggerhead Turtle are at relatively high trophic levels, with the Nile Crocodile also being the largest predator in Africa. We sampled eggs from these three species (27 crocodile, and 10 each from the two turtle species) and analysed the shells and contents separately for metallic elements using ICP-MS. Trophic level, body size, and migratory patterns influenced the concentrations in shells and egg contents, but crocodiles generally seem to have lower concentrations than the sea turtles. Compared with data from elsewhere, sea turtle eggs had lower concentrations, but crocodile eggs had higher copper and mercury concentrations. Comparisons between eggshells and egg content elemental compositions for each species clearly showed that eggshells cannot be used as proxies for egg contents. Sampling therefore, requires the collection and analyses of unhatched eggs. Additional research is needed on the effects of metal concentrations in the eggshells on the hatching process. These refinements allows the RUD values which are then used to recalculate the TWA factor used in the risk assessment. TheTier 1 risk assessment uses default values for residues levels (in terms of residue per unit dose, RUD) and residue decline (in terms of a time weighted average factor, TWA). When the Tier 1 risk assessment indicates a high risk a higher tier assessment is required. One option is to refine the estimate for the level of residues that wildlife consume through their diet by calculating specific RUDs or by deriving substance/crop specific DT50 values which are then used to recalculate the TWA factor used in the risk assessment. Often the residues studied subjects in the risk assessment. Environmental and Marine Studies (CESAM); F. Gonçalves, University of Aveiro / Department of Biology & CESAM, N. Abrantes

MO058 Increasing salinisation effects on Pelophylax perezi populations - Could historical exposure drive current patterns?
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Coastal wetlands are a priority for conservation because they are biodiversity hotspots and have high ecological importance in ecosystem services. However, they are increasingly exposed to strong anthropogenic pressure and climatic extremes. Additionally, Intergovernmental Panel on Climate Change (IPCC) reported alarming projections for sea levels rise in the AR5 at 2013. Within this scenario, it is foreseen the salinisation of low-lying coastal freshwater ecosystems due to the intrusion of seawater, which will adversely affect many populations of amphibians. Pelophylax perezi is distributed along all coastal territory in Portugal, where there are some populations historically exposed to environmental and levels of seawater. Though this species has been reported as tolerant to high salinity levels, there is a lack of knowledge regarding long term effects of salinization to natural populations of this amphibian species. The present work aimed at characterizing the lethal and sublethal sensitivity to salinization of early life stages of P. perezi originating from reference and salinized natural populations. Embryos (Gosner stage h-8) were exposed for 60h, and to what concerns to the input of power to several dilutions of seawater and concentrations of NaCl (used as a surrogate of seawater to increase salinity). The following endpoints were monitored: time until hatching, growth and feeding. Comparing the results for time to hatch between salinization-impaired and non-impaired populations, differences were not found. For this endpoint, NaCl revealed to be more toxic than seawater (EC50 of 14.04 and EC50=1.19 mM/cm² for salt and NaCl, respectively). However, for the sub-lethally monitored endpoints (tadpole growth, weight and feeding) NaCl caused significant adverse effects. Highly diluted seawater increase growth on tadpoles from salinization-impaired population, but for non-impaired populations, tadpoles’ growth decrease with the decrease of seawater dilutions.

MO059 Wildfires effects on aquatic invertebrates organisms with in situ bioassays
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In the last decades, the increased frequency and extent of wildfires have become a societal and environmental problem in Portugal. Among the distinct environmental impacts, the role of wildfire on the water quality has increasingly received research attention, particularly in what concerns to the input of polycyclic aromatic hydrocarbons (PAHs) and metals associated to ashes. However, their toxic effects on the aquatic life have been largely ignored. The main goal of this study was to assess the off-site effects of wildfire on freshwater organisms through the use of in situ bioassays. These bioassays were conducted in a recently burned eucalyptus area located in Préstimo (Âgueda, central Portugal) and occurred after the first wildfires in the region in the mid nineties. In the mid nineties, the same area was burned during two in tributary streams. In the river, two sites were chosen, one located upstream of the burned area (RUS-upstream river) and another located downstream, within the burned area (RDS-River downstream). The same was done for the tributaries (SUS-stream upstream and SDS-stream downstream in the burned area). Distinct freshwater organisms, including the shrimp Atyaphyta desmaresti (water column organism), the amphipod Echinogammarus meridionalis (water-sediment interface organism) and the benthic insect larvae of Chironomus riparius were exposed in all four sites, using dedicated test chambers. After two days of field exposition, the mortality and post-exposure feeding inhibition were evaluated. The lethality was not sensitive to discern impacts among the assessed sites because the results showed negligible mortality for all the species and sites. Conversely, the sub-lethal post-exposure feeding inhibition, revealed a decrease in the feeding rate of organisms from the sites impacted by wildfire (RDS and SDS). Unlike, the sites outside the burnt area (RUS and SUS), showed no adverse effects in this endpoint. These results highlight that in situ bioassays are a suitable tool to assess the risks of wildfire to aquatic species and that the post-fire runoff, rich in substances such as PAHs and metals, can sub-lethally impair the aquatic organisms in water bodies located downstream the burnt area.

MO060 Estricogenic effects of an Organophosphorous Flame Retardant (TCPP) on Edible Sea Urchin “Paracentrotus lividus”
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Abstracts

New synthetic chemical compounds, like Organophosphorous Flame Retardants (OPFRs), are widely used by the industry as plastic additives in common life objects, or overlaying woods, fabrics, etc. They pose environmental risk due to their effects as androgenic or estrogenic endocrine disruptors, imitating male or female hormones respectively, interfering in vital functions of the organism. Increasing presence of those chemicals in the water due to the incomplete elimination in the wastewater treatment plants is emerging as a new problem in water contamination. OPFRs, like Tris (2-Chloro-1-Methyl-1-propyl) Phosphate (TCP), are a typical flame retardant in plastics, being the most detected chemical in the aquatic system. Possible toxic effect of this chemical has not been deeply evaluated yet. This study aims to explore the possible effect of TCP as an endocrine disruptor on the edible sea urchin Paracentrotus lividus. 392 individuals have been used. The analysis of total DNA damage. Behavioral responses to TCP and TEQs were analyzed at 7 and 28 days. TCP exposure did not cause histological damages in the gonads, and the bioaccumulation in the tissues was moderate (mean BCF=28 L/Kg WW). However, the results of the GI in this study, support the idea of an endocrine disruption action of TCP in females exposed to the compound, thus the compound could be catalogued as estrogenic for this marine biological model. Keywords: Edible Sea Urchin, Organophosphorous flame retardant, endocrine disruptor.

MO061

Short-term effects of fluoxetine exposure on biomarker and behavioural responses of an estuarine fish

L.A. Duarte, M.P. Pais, P. Reis-Santos, MARE - Marine and Environmental Sciences Centre; H.N. Cabral, Faculdade de Ciências da Universidade de Lisboa / MARE-FCUL; V.F. Fonseca, MARE Marine and Environmental Sciences Centre Pharmaceutical compounds are routinely discharged into the aquatic environment. There is growing concern whether they elicit deleterious effects on aquatic organisms, following point source acute exposure as well as chronic exposure. Aquatic contaminants like fluoxetine are frequently detected in both freshwater and coastal systems and have deleterious biological effects at very low concentrations. Nonetheless, contradicting evidence has been reported with lack of consistency in responses across studies. In this context, short-term effects of fluoxetine exposure were analysed in common goby Pomatoschistus microps, an estuarine resident species. Two experiments were conducted: where 1) fish were exposed to environmentally concentrations of fluoxetine for 96h (0.1 - 100 μg/L) and 2) fish were exposed to high fluoxetine concentrations for 1h (1, 5 and 10 μg/L). Acute toxicity was assessed via multiple biomarker responses, namely antioxidant enzymes activity, detoxification enzymes activity, neurotoxicity and biomarkers of deleterious effects (e.g. catalase, glutathione S-transferase, acetyleholinesterase, lipoxidation, lipid peroxidation). Behavioral responses also examined, concerning activity and feeding behaviour. Overall, results provided insights into the effects of fluoxetine at the sub-individual and individual-level responses in this species and are discussed concerning realistic exposure scenarios as well as their potential implications to estuarine populations.

MO062

Assessment of PCDD/Fs, dioxin-like PCBs and PBDEs in Mediterranean striped dolphins

F. Capanni, University of Trieste / Department of Life Sciences; J. Muñoz-Arnanz, IQOQ-CSIC / Department of Instrumental Analysis and Environmental Chemistry; L. Marsili, M. Fossi, University of Siena / Department of Physical Sciences, Earth and Environment; B. Jimenez, IQOQ-CSIC / Department of Instrumental Analysis and Environmental Chemistry Marine mammals are exposed to a variety of persistent organic pollutants (POPs) that bioaccumulate in marine ecosystems. In the present study, blubber samples from ten stranded Mediterranean striped dolphins (Stenella coeruleoalba) were used to investigate levels of polychlorinated dibenzo-p-dioxins and furans (PCDD/Fs), dioxin-like polychlorinated biphenyls (DL-PCBs) and polychlorinated diphenyl ethers (PBDEs) using gas chromatograph coupled to a high-resolution mass spectrometer and by using the automated sample preparation system. Samples were analyzed for PCDD/Fs which were found in an order of magnitude lower. Yet, they were generally much higher that those reported for sperm whales from the Sea of Cortez and from Australia. Regarding PBDEs levels, our results were lower that those reported for sperm whales from North-Antarctic. The PCDF congener profile (hexa>hepta>octa>nona>deca) was relatively similar to those reported for sperm whales from Australia and to those reported in blubber of striped dolphin (Stenella coeruleoalba) from the Mediterranean Sea. In contrast, the PCDD congener profile (hexa>penta>hepta>octa) was quite different from those with a lower contribution of higher chlorinated congeners and a higher contribution of lower chlorinated congeners. Total calculated TEQs ranged from 275 to 987 pg g⁻¹ lw and was greater than threshold levels of 210-280 pg g⁻¹ lw, in all blubber from the Mediterranean Sea (15713-75560 pg g⁻¹ lw). This concentration values obtained were 6120 ng g⁻¹ (210-20800 ng g⁻¹) for DL-PCBs, 612 ng g⁻¹ (312-1390 ng g⁻¹) for PBDEs and 57.8 pg g⁻¹ (45.8-83.5 pg g⁻¹) for PCDD/Fs.

MO063

Biochemical and molecular responses to organic contaminants in bottlenose dolphins (Tursiops truncatus geyreaux) from southern Brazil.

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Vitamin A (VA) and its avitaminosis associated to exposure to persistent organic pollutants (POPs) in cetaceans. Such effects threaten the maintenance of cetacean populations, emphasizing the need for biomarkers that indicate early-on biological responses to POPs. The present work evaluated biomarker response to organic contaminants in bottlenose dolphins subspecies geyreaux from two estuarine systems of southern Brazil impacted by agricultural and industrial runoff: Laguna Estuarine System (LES) (n=7) and Patos Lagoon Estuary (PLE) (n=10). Antioxidant enzymes and mRNA transcript levels of genes related to xenobiotic detoxification (AhR, ARNT, CYP1A, GST, MT2), antioxidant defense (GST-π, GPx 4, GR) and immune response (IL-1, MHC-II) were analyzed in integument samples obtained through remote biopsy. POPs were measured in the blubber of the same animals. Generalized linear models (GLMs) were used to analyze the response of each biomarker to PCBs, DDTs, Mirex, Chlorodanes (CHL), Hexachlorobenzene (HCB), sampling season (winter or summer) and location (LES and PLE). The best model to describe each biomarker response, with the lowest Akaike Information Criterion (AIC), was chosen using backward selection. GLMs results indicate that...
the transcript levels of all studied genes were higher in winter when compared to summer, potentially due to enhanced metabolism over colder months. mRNA transcript levels of AHR, GR, IL1 and MT2 genes correlated positively with increasing levels of blubber 2PCBs, supporting the occurrence of biological response to this class of contaminants. GLMs for MT2 indicated that the transcript levels of this gene are higher in dolphins from LES, possibly due to greater metal inputs in this area. GR activity was higher in dolphins with higher fecal TC and SE concentrations, indicating the skin of bottlenose dolphins is altered due to exposure to 2PCBs and 2PDBEs, which co-occurred with 2PCBs and Mirex. Absence of influence of other contaminant classes over biomarker response might be due to low contaminant levels in sampled dolphins. This influence might also have been overshadowed by the effects sampling season. Nonetheless, results indicate a significantly high exposure to PCPs, PCBs, and Mirex to trigger a biological response in dolphins from these small resident coastal populations, particularly susceptible to the negative effects associated to contaminants.

MO065 Monitoring Eleonora’s falcon conservation status both at its breeding and non-breeding grounds, using biological (stress indices) and environmental data
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The present study investigated a battery of stress indices in blood and liver of Eleonora’s falcon (Falco eleonorae Géné, 1839), a colonial breeding raptor of the Mediterranean that overwinters in SE Africa, mainly in Madagascar. In particular, cholinesterase (ChE), acetylcholinesterase (AChE) and butyrylcholinesterase (BChE) activity, as well as cellular abnormalities (MN assay) were measured in blood samples collected from wild individuals, captured on the island of Antikythira (Greece), in May (N=13) and September 2017 (N=19). The results derived from the samples that were collected in May are indicative of the habitat quality at the species’ wintering and/or staging areas, while the ones derived from the samples collected in September are indicative of the habitat quality at the species’ breeding grounds. Moreover, in order to investigate the water quality in the breeding area of F. eleonorae, natural water pond samples were collected in September 2017 and further analyzed for the presence of heavy metals. Additionally, heavy metals were measured in liver of an individual found dead near the water ponds. According to the results, total plasma ChE activity ranged between 3.370±0.433 - 11.34±0.829 nmol m-1 min-1 in May and 1.44±0.079- 9.31±1.618 nmol m-1 min-1 in September. AChE activity remained almost constant between the two sampling periods, while BChE in May was significantly higher than September. Total nuclear abnormalities showed almost similar frequencies in both periods, while cytoplasmic abnormalities observed in September were significantly higher than in May. Cr and Cu levels in water samples were found to be within natural levels, while Cd and Pb concentrations were lower than the MAC-EQS values. All heavy metals were significantly lower than the MAC-EQS values. However, no significant differences are observed between the water quality of the breeding area and the non-breeding area. The results of the present study showed for the first time that the assessment of a battery of stress indices in tissues of F. eleonorae, together with chemical analysis of data derived from their natural habitats, could serve as a valuable tool for elucidating the quality of its foraging grounds and, hence, the impact of land use on the species’ conservation status.

MO066 Optimising design and analysis of acute effect field studies
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Vertebrate risks assessments of a plant protection product (PPP) may indicate an acute risk to wild birds and mammals and predict effects on population development. This might be driven by (too) conservative assumptions on the exposure side of the equation for the risk evaluation, due to the lack of better data. It is therefore worthy to study deeply the presence of effects in the field in order to obtain a proper risk assessment. We highlight three complementary ways to improve the quality of such findings. An optimised study design can comprise the so-called ‘extensive approach’, by using a great area or number of agricultural fields in different study sites, with the ‘intensive approach’ by using radio-tracking techniques in a control/treatment design. This double approach covers the natural variation in parameter estimates and enables the identification of possible treatment effects. The radio-tracking technique is sensitive enough to monitor the fate of individuals in a treated population over a long period of time, and to find out if they reach mortality. However, in most cases the critical point is the disappearance of individuals; an increase in their number can indicate a greater vulnerability to other stressors. In the context of a good study design, we also propose an improved statistical evaluation to increase the detectability of effects in comparison to earlier studies. The Kaplan-Meier survival curve and the Cox proportional hazards model are recommended as methods for the analysis of survival information. The Cox model is a well-known statistical technique used in medical tests. It provides an estimate of the treatment effect on survival adjusted for other explanatory variables. Moreover, as it isolates the effects of treatment from the effects of other covariates, the assessment of results of such field studies is facilitated. Additionally, an essential part of every statistical evaluation is to know the minimum number of individuals needed in order to perceive actual treatment effects in the statistical output. Using data from generic radio telemetry studies on real untreated populations of wood mouse and several bird species, we run simulations of acute effects for different scenarios. The results show that minimum sample size is highly dependent on, first, the species, and second, the action mode and persistence of residues of each specific PPP.

MO067 Assessing impacts of legacy pollutants on wildlife of the Trinity River (Texas, USA) using Neotropical Cormorants as indicator species
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The Trinity River (Texas, USA) has been historically known as a polluted river because of its proximity to the Dallas-Fort Worth area and also because of known discharges of sewage and agricultural irrigation waters to the river. Surprisingly, there are no studies regarding the presence of legacy contaminants in the river and their impacts to wildlife. The objectives of this study were to determine accumulation and potential impacts of persistent organic pollutants such as organochlorine pesticides, PCBs, and PBDEs on nesting aquatic birds of the Trinity River, using Neotropical cormorants (Phalacrocorax brasiliensis) as indicator species. Adult and first year cormorants were collected from two sites on the Trinity River Watershed during 2014 and 2015. Tissue sections from liver, spleen, kidneys, and gonads, were used for histopathology analysis, and a portion of the liver was analyzed for OC pesticides, PCBs, and PBDEs. Results show that the Trinity River is contaminated with a wide range of OC pesticides, PCBs, and PBDEs. Concentrations of PCBs, PBDEs, and Hg. Surprisingly, all the contaminants were present at low concentrations and below those that could be associated with adverse effects; however, altered structure, composition and function, were detected in the livers and kidneys of most samples. A novel coccidian Eimeria sp. was also detected in the kidneys of several cormorants. Our results suggest that aquatic birds using the Trinity River watershed and nearby fields may be early susceptible to the negative effects associated to PCBs, pesticides, PCBs, and PBDEs. These results should be useful for wildlife managers regarding concerns over contaminant impacts of the Trinity River on wildlife.

MO068 Testing the effects of a neonicotinoid insecticide on songbird migration
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Recent decades have seen a dramatic increase in the application of neonicotinoid insecticides, which are now the most widely used class of insecticides in the world. Migratory seed-eating birds that use agricultural landscapes for refueling may be directly exposed to OC pesticides and may experience toxic effects to their reproductive, digestive, and/or immune systems. To determine the effects of a neonicotinoid insecticide on songbird migration to test the direct effects of imidacloprid on the migratory behavior of seed-eating passerines. In a previous captive study, we found birds exposed to imidacloprid exhibited no differences in general behavior and feeding rate compared to control birds. In a second study, we observed a significant mass loss and stopped orienting correctly in behavioural trials, whereas control birds maintained body mass and a seasonally appropriate northward orientation. To corroborate results from captive trials on free living birds, we conducted a study on radiotagged white-crowned sparrows following a single oral dose of imidacloprid. Birds were caught in Ontario, Canada during spring migratory stopover and exposed to imidacloprid via gavage to a number of birds with either 1.2 mg/kg bw or 3.9 mg/kg bw (n = 12 birds/treatment). Birds were held for approximately 6 hours, and their body mass and food intake were monitored. Individuals were then tagged with uniquely coded transmitters and released into into a MOTUS array of automated telemetry towers in Southern Ontario, Canada to track their movements on a landscape scale. We found that high dose birds significantly reduced food consumption, and imidacloprid exposed groups lost a significant amount of body mass relative to controls within 6 hours of dosing. Using automated telemetry data, we found that birds with the longest stopover durations were in the imidacloprid treated groups. Further analysis of the telemetry data is being used to determine effects on speed of travel and direction of migratory movements across a large water barrier.

MO069 A synthesis of the interactions between anticoagulant rodenticides and wildlife
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Anticoagulant rodenticides (ARs) are the mainstay of rodent control throughout the world. Regulatory risk assessments indicate ARs pose a significant risk to non-target wildlife but AR use remains widely authorised because the benefits (particularly to human health) are deemed to outweigh the environmental risks. Recently, an authoritative reference text, prepared by 24 international scientists, reviewed the main issues related to ARs and wildlife, specifically: AR use, regulation, exposure pathways, toxicity, mechanism of action, pathology, pharmacokinetics, genetic resistance, non-target risk and its mitigation, alternatives
to ARs and integrated pest management (IPM). Broad concepts that emerged were: there is high conservation of the blood clotting process and so ARs can affect a wide range of non-target species; development of genetic resistance in target species led to global use of the more acutely toxic and persistent second-generation ARs (SGARs); vitamin K1 can be an effective antidote (unlike for many rodenticides); variation in non-target sensitivity may be due to pharmacokinetic, ecological and behavioural processes; >50% of predatory species contain AR residues; AR residues are already present in exposed, food webs and of AR-induced mortality requires additional evidence (e.g., clinical signs, haemorrhagic lesions); probability of death in relation to AR residues may help assess extent of mortality in populations; tissue residues are informative of exposure but dietary AR concentrations are more suited to assess risk; and primary AR exposure associated with recreational activities may be considered of non-target species population declines, but there is no clear evidence for secondary AR exposure causing population declines. Alternatives to ARs (e.g., bromethalin, cholecalciferol, zinc phosphide) also pose a significant risk to non-target wildlife, livestock, companion animals and people. There are a number of key information needs to better understand the environmental risk from ARs. These are: improved understanding of ARs’ effects; dose limits for conservation risks; knowledge of exposure and effects in invertebrates and lower vertebrates; enhancement of resistance management in target species; development of safe alternative chemical and non-chemical methods. It is anticipated that addressing these research priorities would reduce the potential AR-related conflicts between protection of human health and wildlife.

MO070 Anticoagulant rodenticides in red kites (Milvus milvus) in Britain


Second generation anticoagulant rodenticides (SGARs) can be toxic to all mammals and birds. Studies have shown that, in Britain, there is widespread exposure to SGARs in a diverse range of predatory mammals and birds, including red kites (Milvus milvus). This species may be particularly at risk as it scavenges dead rats, a target species for rodent control. Investigation of SGAR exposure in red kites across Britain from independent projects can be facilitated via the WILDCOMS network (www.wildcoms.org.uk/) has facilitated collation of these data sources and a resultant national-scale annual assessment of exposure in red kites. We report the findings of the first such national scale national exposure which was conducted for red kites found dead in 2015. Carcasses were typically found by members of the public, sent to an investigating laboratory, necropsied and analysed for liver SGARs by Liquid Chromatography Mass Spectrometry. Of 26 red kites from England & Wales that were analysed, all had detectable residues of difenacoum and brodifacoum; most also contained bromadiolone. Difenacoum was less frequently detected and flocoumafen was not detected in any birds. SGAR concentrations ranged from 50 to 1266 ng/g wet wt. (arithmetic mean: 372 ng/g). Post-mortems indicated that 9 (35%) of the kites had internal hemorrhaging that was associated with detectable trauma; most had elevated sum SGAR liver concentrations. On the basis of these two factors, it is considered probable that SGARs were a contributory cause of death in these birds. Residue data were also available for 6 red kites from Scotland. Three (50%) had liver residues of at least two SGARs (bromadiolone and difenacoum); brodifacoum was also detected in one of these kites. SGARs were assessed to be a contributory cause of death in the bird that had residues of three SGARs. The data for Scotland, although limited, suggest that exposure of red kites in 2015 may have been less marked than in England & Wales, as has been found for other species collected in other years. Overall, these results suggest all red kites in England & Wales are at least exposed to SGARs and that poisonings are not uncommon. Despite this, the red kite population has greatly expanded in Britain as birds recolonise former haunts. The extent to which exposure to SGARs may affect future population growth merits further investigation.

MO071 Environmental determinants of the exposure to anticoagulant rodenticides in non-target species

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Anticoagulant rodenticides have some similarities with other bioaccumulative persistent organic compounds, because of their frequent presence in many predatory species. In addition, the fact of being highly toxic substances makes this biacumulation particularly harmful for these predators. Considering that the use of rodenticides occurs mainly in areas with high density of rodents that are in turn prey to multiple predators, we can also expect an ecological trap scenario. We studied the levels of second generation anticoagulant rodenticides (SGARs) and the environmental factors that influence such exposure in non-target species. The analysis included liver samples of wild animals (n=244) found dead between 2007 and 2015 in the region of Valencia (Spain). The sampling included 25 small mammals (11 reptile, 16 mammals and 32 birds). Liver samples were obtained by LC-MS and the presence of SGARs was statistically analysed with generalized linear models with a binary logistic response to study the effect of environmental or habitat characteristics including human population and livestock density and types and surface of crops. SGARs residues were detected in 83 (34%) of the analysed animals. Among small mammals, corresponding to 25 (51%) species. Ten species (53 individuals, corresponding to four mammals and six birds, had residues >200 ng/g, which is the threshold associated with adverse effects: these included common raven (67%), red fox (50%), red kite (38%), eagle owl (25%), stone martens (23%), common buzzard (17%), Western marsh-harrier (17%) and Eurasian badger (14%). The spatial analysis at the municipality level has allowed to identify the percentage of municipality urban land. The ecological Centre; A. Covaci, University of Antwerp, Toxicological Centre / Toxicological Centre Dep of Pharmaceutical Sciences; D. Herze, NILU Norwegian Institute for Air Research; B. Styhrslave, University of Copenhagen / Section of Analytical Biosciences Department of Pharmacy; V. Jaspers, Norwegian University of Science & Technology / Biology

The international research project NewRaptor (ID 230465/F20, funded by the Norwegian Research Council and the Norwegian University of Science and Technology) aims to investigate the exposure and effects of emerging chemicals in birds of prey. The raptores under investigation include the terrestrial Northern goshawk (NG - Accipiter gentilis) and the marine White-tailed eagle (WTE - Haliaeetus albicilla) from Norway and NG from Spain. During the breeding seasons of 2015 and 2016, blood and body feathers were obtained from the chicks (H. albus) and from paternal parents (N = 160 for NG and N = 145 for WTE) for periods of 4-9 weeks old. The samples were analysed for novel brominated flame retardants (nBFRs), organophosphate flame retardants (PFRs) and per- and polyfluoroalkyl substances (PFASs), along with trace elements and legacy persistent organic pollutants (POPs). Significant differences were found between the two species (with WTE generally showing higher levels of pollutants), but also within species, depending on the location. PFASs were generally found at the highest concentrations, with perfluorooctane sulonate (PFOS) being the most important component. nBFRs and PFRs were found at very low or non-detectable levels in blood plasma. Further, pollutant effects on different biochemical, immunological and endocrine parameters were assessed. We have performed controlled in vivo experiments, studies in Japanese quail (Coturnix japonica) and chicken (Gallus domesticus) as model species, both with single compounds and in a mixture scenario. The compounds for the quail exposure study included Dechlorane Plus (DP), tris(1,3-dichloro-2-propyl) phosphate (TDCIPP) and their 1:1 mixture, while PFOs, F-53B (PFOS replacement product) and their 1:1 mixture were used in chickens. Effects on gene expression and activity of anti-oxidative enzymes (catalase, superoxide dismutase and glutathione peroxidase), lipid - and protein oxidative damage and biotransformation (cytochrome P450). We investigated further, the biological analysis of corticosterone and progesterone was performed using HPLC-MS/MS. Gene expression and enzyme assays on similar endpoints will be performed on NG samples in January 2018 and will be presented alongside the results from the in vivo exposure studies at SETAC. This will enable discussing the potential usefulness and pitfalls when extrapolating from laboratory dosing studies using model species to field assessments in raptors.
The potential of feathers as a biomonitoring tool for fluoxetine in wild birds
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The antidepressant fluoxetine has been identified as a contaminant which may pose a risk to wild birds. However there is little empirical evidence regarding which bird species are most at risk of exposure to fluoxetine, in terms of concentration levels in wild bird samples. A significant barrier to sampling wild birds is that fluoxetine is cleared very quickly from systemic circulation and plasma concentrations might be expected to fall to levels below the limit of detection in less than one hour post-exposure. Facial sampling presents a similar problem. However, fluoxetine is detectable in the hair of humans and other mammals. We hypothesised that fluoxetine might likewise be detectable in feathers. Moult occurs over a period ranging from weeks to months and during this time, growing feathers have a blood supply. If a wild bird was exposed to fluoxetine during this period, the compound could be laid down in the feathers and subsequently be detectable. We conducted an avian study with wild caught Eurasian starling (Sturnus vulgaris) to determine whether fluoxetine administered during a period of feather regrowth is deposited in the feathers in detectable concentrations. We removed two rectrices (tail feathers) from each bird. We then administered a dose of fluoxetine at an environmentally relevant concentration (3.8 μg d−1) each weekday throughout the regrowth period. We plucked the new feathers once they were fully regrown and analysed them by LC-MS/MS for fluoxetine and its major active metabolite, norfluoxetine. Our preliminary results indicate that fluoxetine is detectable in the feathers and we will present information on the concentrations present and whether they are correlated with levels in other tissues such as liver and brain. We discuss the extent to which feathers have potential as tools for monitoring the occurrence and severity of exposure to fluoxetine in wild birds.

MO076 Different approaches comparison for evaluation of hypopharyngeal glands activity: a case study with Honeybees (Apis mellifera L.)

Honeybees (Apis mellifera L.) are beneficial arthropods that play important roles in natural ecosystems, agriculture and in the food and pharmaceutical industries. One of the conditions for maintaining healthy colonies is the proper development of the honeybee workers (hypopharyngeal glands, HPG) which produce proteic substance to feed larvae and queen. The aim of this study was to validate the different algorithms (including obtaining the material) to conduct the hypopharyngeal glands development effect studies, which investigate effects under real use conditions and in different habitats. Population size and development, body weight, reproductive performance, survival rates were determined and analyzed using linear mixed models. Results of the study will be presented in detail in the poster. The results support the fact that field-effect studies, which investigate effects under real use conditions and are included as higher tier refinement option in the EFSA Bird and Mammal Guidance Document (EFSA, 2009), are a useful tool for the higher tier risk assessment, even though no internationally agreed standard protocol for mammal field-effect studies is existing yet.

MO075 Monitoring NSAIDs in carrion and avian scavengers form Spain: preliminary results after diclofenac registration for veterinary use
R. Martín, IREC-CSIC- UCLM / Grupo de Toxicología de Fauna Silvestre; M. Herrero, Instituto de Investigación en Recursos Cinegéticos (IREC); P.R. Camarero, Instituto de Investigación en Recursos Cinegéticos, CSIC-UCLM-JCCM / Grupo de Toxicología de Fauna Silvestre; I.S. Sanchez-Barbudo, UCLM-CSIC / Grupo de Toxicología de Fauna Silvestre; R. Vidal, I. Marchesi, IREC / Grupo de Toxicología de Fauna Silvestre.

The serious impact of diclofenac on Asian vultures raised the alarm of the deficient environmental risk assessment of some veterinary drugs. In the case of diclofenac, there was an evident gap in the knowledge of the high toxicity that this non-steroidal anti-inflammatory drug (NSAID) has in Old-World vultures that can feed on treated livestock (LD₅₀ in Gyps bengalensis of 98-225 µg/kg body mass). Avian scavengers (vultures and black kites) are the focal species suggested for rice in the wet environments of rice paddies. In this work, we studied the presence of NSAIDs in carrion animals (kidney, liver and muscle of pig, n=125) supplied in “muladares” (live food for vultures) to assess the occurrence and severity of exposure to NSAIDs residues in tissues of carrion animals (kidney, liver and muscle of pig, n=125) supplied in “muladares” (live food for vultures). We have also studied the presence of NSAIDs residues in tissues of avian scavengers (vultures and black kites) in 225 individuals sampled in different locations in Spain. Despite the experience with diclofenac presence. An examination of this muscle sample showed a pale area adjacent to a congestive portion that may correspond to the injection point of the diclofenac in the pig. Fluixin was the only NSAID detected in the studied avian scavengers. Two out of 22 Eurasian griffons (Gyps fulvus) analysed had 330 and 23 ng/g of fluixin in liver and muscle tissues of Aegypius monachus 2.83 μg/g of fluixin in liver, but it was diagnosed as an atrophic poisoning at the wildlife rehabilitation center. Lesions in the kidney and visceral gout have not been observed macroscopically or microscopically in 15 Eurasian griffons analysed. Residue levels in carrion and scavengers indicate a limited risk of poisoning, possibly because treated livestock died several hours after NSAID injection. However, veterinary use of NSAIDs can still be a threat for scavengers if veterinarians and farmers ignore the associated risk when treated livestock is used to feed vultures.

MO074 Field-effect studies as a suitable method to assess effects of plant protection products on free-living common voles (Microtus arvalis): A case study with the fungicide iprodione
O. Fuelling, C. Miersch, Tier3 Solutions; S. Steiger, BASF SE, Agrarzentrum Limburg/Idstein.

After foliar spray application of a plant protection product on crops, food sources of small mammals may be potentially contaminated with this product. Ingestion of treated food could possibly lead to effects on the population level (e.g. reproductive impairment etc.). In the presented study, it was examined if there were any long-term effects from repeated foliar spray applications of the fungicide iprodione on populations of the common vole, Microtus arvalis. The field-effect study was conducted in Germany during the main reproductive period of the common vole on 14 commercially used grassland fields. Regular trapping sessions which followed a capture-mark-recapture design were conducted from June to November 2014 on treated and untreated (control) grassland fields, as well as in adjacent habitats. Population size and development, body weight, reproductive performance, and survival rates were determined and analyzed using linear mixed models. Results of the study will be presented in detail in the poster. The results support the fact that field-effect studies, which investigate effects under real use conditions and are included as higher tier refinement option in the EFSA Bird and Mammal Guidance Document (EFSA, 2009), are a useful tool for the higher tier risk assessment, even though no internationally agreed standard protocol for mammal field-effect studies is existing yet.

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MO077 Bird and mammal focal species for pesticide risk assessment in rice
M. Valioglu, C. Dietzen, S. Laucht, F. Sotti, J. Ludwigs, Rifcon GmbH

Ecotoxicological risk assessment for birds and mammals is required for the registration of pesticides in Europe to assess potential risks to wildlife through the intake of contaminated diet items. To ensure a realistic and reliable risk assessment, bird and mammal focal species should be used that are representative for the species actually occurring in the crop of concern. However, in the relevant guidance document on bird and mammal risk assessment by the European Food Safety Authority (EFSA), rice is to date pooled with other cereals such as wheat and barley, despite the obvious peculiarity of rice cultivation. The generic focal species suggested for rice are thus those known from the dry environments of cereal fields. To address this issue, we conducted a comprehensive literature review on bird and mammal species regularly occurring in rice paddies at the relevant time periods of pesticide exposure to identify appropriate focal species candidates for risk assessment. Our results show that the relevant species occurring in the wet environments of rice paddies indeed clearly differ from the focal species suggested for rice risk assessment for wildlife in cereals and thus provide a baseline for more realistic and risk-specific risk assessment for birds and mammals.

MO078

161 SETAC Europe 28th Annual Meeting Abstract Book
Non-invasive assessment by feathers of lead exposure and its relationship with stress hormones in bearded vultures from the Alps

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A re-introduction project of bearded vulture or lammergeier (Gypaetus barbatus) has been carried out in the Alps since 1987. There are several factors that can affect the survival and post-mortal use of feathers, such as feathers moulted by birds. The objectives of this study are to quantify the degree of exposure to lead that the bearded vultures have in the Alps post mortem feathers found in the field, relating them to corticosterone levels and defining the factors that affect the fluctuation of these two components along transverse segments of feathers. The analysis of the segments of 20 different feathers from different bearded vultures revealed that the abnormal exposure to lead (probably due to the ingestion of ammunition) had a prevalence of 15% (three feathers with levels above 2 μg/g of Pb in rachis), and that the annual incidence of such exposure was 30% (a feather with an abnormal exposure during its development of approximately two months). The concentration of lead in bearded vulture feathers was stable for 90% of the feathers along with following blood clinical chemical parameters (BCCPs): albumin, total proteins, aspartate aminotransferase, alanine aminotransferase, creatine kinase, amylase, glucose, creatinine, bilirubin, potassium and uric acid. Stable carbon (δ13C) and nitrogen isotopes (δ15N) were analysed in feathers to evaluate inter-and intra-specific contaminant exposure. Due to the low amount of feather samples, Hg and its effects at the biochemical/physiological level in White-tailed eagles (Haliaeetus albicilla) and Northern goshawks (Accipiter gentilis) nestlings from Norway

P. Gómez-Ramírez, University of Murcia / Department of Toxicology; J.O. Bustnes, Norwegian Institute for Nature Research / Fram Centre; G.S. Eggem, Norwegian University of Science and Technology / Biology; I. Eulaers, University of Antwerp / Biology; F.C. Bjoern, University of Tromso / Department of Oceanology; J.M. Pérez-García, University of Leida / Department of Animal Sciences; T.V. Johnsen, Norwegian Institute for Nature Research NINA; A. García-Fernandez, University of Murcia / Sociosanitary Sciences; V. Jaspers, Norwegian University of Science & Technology / Biology

Assuring the effectiveness of such restrictions and the current levels of these chemicals in the environment, biomonitoring using birds is very useful. Hg has caused detrimental effects in birds such as haemotoxicity, immunotoxicity and endocrine disruption e.g. suppression of baseline corticosterone. The aim of this study was to assess the exposure to Hg and its effects at the biochemical/physiological level in White-tailed eagles (Haliaeetus albicilla) in the Alps by analysing moulted feathers found in the field, relating them to the concentration of corticosterone, and was significantly influenced by the collection area and the individual (feather). The present study we observed that exposure to elevated levels of lead can occur in a high percentage of individuals in a population throughout the year, which in the case of the lammergeier of the Alps can entail a risk to the sustainability of the population if this exposure reach lethal levels.

MO079 Post mortem stability of phase I and II biotransformation enzymes in the liver of kelp gull Larus dominicanus

J.F. Mattos, Universidade Federal de Santa Catarina / NEPAQ, Departamento de Agricultura; D.D. Lima, Universidade Federal de Santa Catarina / Biogasuniq; B. Righetti, Universidade Federal de Santa Catarina / NEPAQ-CCA; L.O. Villas Boas, V.H. Dias, Universidade Federal de Santa Catarina / CCB; S. Costa-Silva, C. Kolesnikovas, M. Antonelli, Associação R3 Animal; J. Ramírez, UAB

The measurement of biomarker responses to chemical contaminants in wild organisms represents a powerful tool in environmental monitoring programs. However, getting biological samples suitable for biomarker analysis may be challenging, since some programs rely on samples collected from carcasses. Shorter periods from death to analysis of biochemical biomarkers provides more accurate results, but how these parameters change at longer post mortem intervals remains unclear. This study involved the post mortem stability of key biotransformation enzymes (glutathione S-transferase, GST and 7-ethoxyresorufin-O-deethylase, EROD) in the liver of kelp gull Larus dominicanus for periods up to 24 hours. Liver tissue of two euthanized animals was sectioned into several 1-cm cubes and stored in individual closed tubes at 25°C for 0, 1, 2, 3, 6, 12, 18 and 24 hours post mortem before liquid nitrogen freezing. Cytosolic and microsomal fractions were obtained from 150 mg of each sample individually and used for GST and EROD measurements, respectively. GST activity proved to be stable after 24 hours (85-90% of initial activity). EROD activity decreased abruptly after the first hour post mortem for both animals. After 3 hours post mortem activity presented down to 71% of initial activity and 28 to 50% of its initial activity after 6h, showing an exponential decrease along post mortem period. Our results indicate that time elapsed since death until sample collection plays an essential role for biotransformation enzymes, especially concerning EROD activity. GST seemed to be more resistant to degradation over time than EROD activity and 28 to 50% of its initial activity after 6h, showing an exponential decrease along transverse segments of feathers. The analysis of the segments of 20 different feathers from different bearded vultures revealed that the abnormal exposure to lead (probably due to the ingestion of ammunition) had a prevalence of 15% (three feathers with levels above 2 μg/g of Pb in rachis), and that the annual incidence of such exposure was 30% (a feather with an abnormal exposure during its development of approximately two months). The concentration of lead in bearded vulture feathers was stable for 90% of the feathers along with following blood clinical chemical parameters (BCCPs): albumin, total proteins, aspartate aminotransferase, alanine aminotransferase, creatine kinase, amylase, glucose, creatinine, bilirubin, potassium and uric acid. Stable carbon (δ13C) and nitrogen isotopes (δ15N) were analysed in feathers to evaluate inter-and intra-specific contaminant exposure. Due to the low amount of feather samples, Hg and its effects at the biochemical/physiological level in White-tailed eagles (Haliaeetus albicilla) and Northern goshawks (Accipiter gentilis) nestlings from Norway.

MO080 Assessment of exposure and effects of Hg levels in feathers of White-tailed eagles (Haliaeetus albicilla) and Northern goshawks (Accipiter gentilis) nestlings from Norway

P. Gómez-Ramírez, University of Murcia / Department of Toxicology; J.O. Bustnes, Norwegian Institute for Nature Research / Fram Centre; G.S. Eggem, Norwegian University of Science and Technology / Biology; I. Eulaers, University of Antwerp / Biology; F.C. Bjoern, University of Tromso / Department of Oceanology; J.M. Pérez-García, University of Leida / Department of Animal Sciences; T.V. Johnsen, Norwegian Institute for Nature Research NINA; A. García-Fernandez, University of Murcia / Sociosanitary Sciences; V. Jaspers, Norwegian University of Science & Technology / Biology

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MO081 Investigating thyroid disrupting effects of organohalogenated contaminants in White-tailed eagle nestlings

M. Lozano, IREC Instituto de Investigación en Recursos Cinegéticos; L.O. Villas Boas, V.H. Dias, Universidade Federal de Santa Catarina / CCB; S. Costa-Silva, C. Kolesnikovas, M. Antonelli, Associação R3 Animal; J. Ramírez, UAB

The measurement of biomarker responses to chemical contaminants in wild organisms represents a powerful tool in environmental monitoring programs. However, getting biological samples suitable for biomarker analysis may be challenging, since some programs rely on samples collected from carcasses. Shorter periods from death to analysis of biochemical biomarkers provides more accurate results, but how these parameters change at longer post mortem intervals remains unclear. This study involved the post mortem stability of key biotransformation enzymes (glutathione S-transferase, GST and 7-ethoxyresorufin-O-deethylase, EROD) in the liver of kelp gull Larus dominicanus for periods up to 24 hours. Liver tissue of two euthanized animals was sectioned into several 1-cm cubes and stored in individual closed tubes at 25°C for 0, 1, 2, 3, 6, 12, 18 and 24 hours post mortem before liquid nitrogen freezing. Cytosolic and microsomal fractions were obtained from 150 mg of each sample individually and used for GST and EROD measurements, respectively. GST activity proved to be stable after 24 hours (85-90% of initial activity). EROD activity decreased abruptly after the first hour post mortem for both animals. After 3 hours post mortem activity presented down to 71% of initial activity and 28 to 50% of its initial activity after 6h, showing an exponential decrease along post mortem period. Our results indicate that time elapsed since death until sample collection plays an essential role for biotransformation enzymes, especially concerning EROD activity. GST seemed to be more resistant to degradation over time, and it thus appears possible to make valid GST activity measurement in selected post mortem liver tissue of kelp gull. Overall, our findings demonstrate that caution is warranted in monitoring programs when comparing biological samples with different intervals between collection and analysis procedures.

MO082 Thyroid-related gene expression, hormones, and thyroid gland histology in American kestrels exposed in ovo to two persistent organic pollutants, SCCPs and TBBPA-BDBPE

A. MacLeod, University of Maryland, College Park / Environmental Science and Technology; P.F. Henry, U.S. Geological Survey / Patuxent Wildlife Research Center; K.J. Fernie, Environment & Climate Change Canada / Ecotoxicology and Wildlife Health; N.K. Karouna-Renier, USGS Patuxent Wildlife Research Center /
Patuxent Wildlife Research Center

Highly brominated flame retardants are being replaced by alternative flame retardants such as Tetrabromobisphenol A bis[2,3-dibromopropyl ether] (TBBPA-BDBPE). TBBPA-BDBPE was introduced as a possible substitute for decabromodiphenyl ether (decaBDE), but has shown similar persistence and environmental transport mechanisms. This additive flame retardant is used in plastic products, textiles, and household electronics. Although it is produced only in the US, Israel, and China, TBBPA-BDBPE is detected in environmental samples and wildlife tissues from across the globe. Short-Chain Chlorinated Paraffins (SCCPs) are priority emerging persistent organic pollutants (POPs) identified as chemicals of concern by the Stockholm Convention, Environment and Climate Change Canada, and the US. Environmental Protection Agency. SCCPs are used in metal lubricants and coolants in metal cutting, and as plasticizers and flame retardants in plastics and paints. SCCPs are of concern because they bioaccumulate in wildlife and humans, are environmentally persistent, transported globally, and are toxic to aquatic organisms at low concentrations.

However, few data are available on the potential adverse effects of TBBPA-BDBPE and SCCPs in birds. A comparative exposure assessment of these two classes of flame retardants was conducted using egg injections in a non-model species, the American kestrel (Falco sparverius) to assess survival, molecular, biochemical, and endocrine, growth and reproductive endpoints. Analyses conducted on tissue samples from hatchlings included thyroid related gene expression in the liver, thyroid hormone levels, and thyroid gland histology. Preliminary results will be presented from the molecular to biochemical to cellular level.

MO083

Bioaccumulation of metals in bats: non-lethal vs lethal sampling to assess risk

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More than 22% of bat species are threatened according to the World Conservation Union. Among the reasons responsible for this decline are the environmental changes due to anthropogenic factors, namely habitat loss through agriculture, forestry, urbanization and industrialization, contamination by pesticides and metals, changes in water quality. There is a growing concern about the possible consequences of environmental contamination in several bat species. Until now, many of the studies on the effects of contaminants in bats are focused on organic contaminants, and the consequences of exposure to other substances (particularly metals), remaining largely unknown. The aim of this study was to evaluate the potential risk of metal contamination in bat species occurring in Portugal, and to evaluate the suitability of non-lethal sampling methods. The concentration of As, Cd, Co, Cr, Cu, Mn, Ni, Pb, Se and Zn was measured in two categories of biological samples (lethal-samples: liver, heart, bone and brain; and non-lethal samples: wing membrane and fur) collected from bat carcasses of four different species (Hypsugo savi, Nyctalus leisleri, Pipistrellus pipistrellus, Pipistrellus pygmaeus). Concerning the metal contamination obtained in each sampling tissue, significant differences were found between the concentrations obtained in each species for all the metals (P < 0.05), except for Zn (P = 0.223). Significant differences were also found between categories of biological samples (P < 0.001), with the highest concentrations of contaminants in the organs and metals (P < 0.001). Depending on the metal, the organ/tissue that showed the highest concentrations varied, but even so far and wing presented the highest concentrations of most of the metals. These results support the hypothesis that non-lethal samples may be useful for studies on wildlife ecotoxicology, and may help to define a protocol capable of being applied at large-scale, to investigate the risk of metal accumulation for bats. For this purpose, non-lethal samples are the best option, and as demonstrated by this study can yield reliable results. Our results therefore provide valuable insights for development of further studies, aiming to understand the importance of metals as a cause for some of the observed declines in bat populations worldwide.

MO084

Metallic element composition of egg contents and eggshells of the Kelp Gull Larus dominicanus

J.D. van Aswegen, North West University (Potchefstroom Campus) / Unit for Environmental Science and Management; L. Nel, N. Strydom, Nelson Mandela University; H. Browman, North-West University / Unit for Environmental Science and Management

The Swartkops River Estuary near Port Elizabeth, South Africa, is an important recreational, industrial, residential, and ecological asset, but under severe pressure. Seabirds are good indicators of trace elements within their environments. Seabirds tend to feed at different trophic levels at different distances from the land and they are long-lived. Pollutants that have accumulated in the seabirds can be excreted in various ways from the body, one being deposition into eggs. Sixteen eggs of the Kelp Gull (Larus dominicanus) were analysed using ICP-MS (EPA 3050b method) for 30 trace elements, for both the contents and eggshells. We selected five elements (Cr, Sr, Ti, U, Zn) to compare and assess the absolute and relative compositional patterns in egg contents and shells. Mean concentrations for Cr in eggshells and egg contents were 3.8 and 18 mg/kg dm, for Sr it was 880 and 12 mg/kg dm, for Ti it was 0.00017 and 0.00022 mg/kg dm, for U it was 0.00057 and 0.000084 mg/kg dm, and for Zn it was 2.1 and 62 mg/kg dm, respectively. Of the five elements, only Sr (p = 0.0141) and Ti (p = 0.0013) concentrations showed significant positive regressions between egg contents and eggshells. Chromium and U showed a positive association, but the regressions were not significant. Uranium also showed no association. The mean mercury concentration in the contents was 0.38 mg/kg dm, and the maximum was 2.1 mg/kg dm. The Toxic Reference Value for mercury in bird egg contents is 2 mg/kg dm, indicating concern about this element in the Swartkops River Estuary. Additional toxic implications, as well as comparisons with concentrations in other media will be discussed.

MO085

Heavy metals concentrations in Mediterranean Osprey eggs: variations by location, habitat and egg constituent part

F. Menu, University of Siena / Department of Physical Sciences, Earth and Environment; A. Sforzi, Maremma Natural History Museum, Grosseto; N. Bianchi, University of Siena / Department of Physical, Earth and Environmental Sciences; C. Leonzio, University of Siena / Department of Physical Sciences, Earth and Environment

The osprey (Pandion haliaetus) has been historically used world-wide as a sentinel species for the biomonitoring of selected contaminants for aquatic ecosystems. In spite of this, occasional and fragmentary information are available for the species at the Mediterranean scale, where relict and vulnerable populations exist. In this study, we analyzed heavy metals concentrations in osprey eggs from three different populations of the Mediterranean basin (Corsica, Italy and Balearic Islands). In total, 21 unhatched eggs were collected, over a period spanning from 2005 to 2016. More than 22% of eggs from Cd, Ca, Cd, Cd (Pb) in these eggs were analysed with the aim to: 1) evaluate geographical patterns of for possible identification of inputs at the local scale; 2) evaluate differences in concentrations between samples from different locations (marine environments and wetlands); and 3) to investigate any differences in concentrations among different parts of the egg (i.e., content, membrane and shell). Samples from the Balearic Islands showed higher Hg concentrations (1.4 ± 1.2 mg/kg on dry weight basis) compared to other samples. Egg shells from marine environments (Corsica and Balearics) had five times greater [Hg] than those from wetlands. Egg content and membrane showed higher Hg concentration values than those of the shell. On the contrary, for Cd and Pb (though with minor differences) higher concentrations were found in the egg shell. Our study represents a framework for each study at regional scale and provides a first set of data for the long-term biomonitoring of heavy metals for the vulnerable osprey populations of the Mediterranean basin.

MO086

Interactive effects of vitamin E and BDE-47 yolk supplementation on morphological and oxidative status of yellow-legged gull embryos

M. Polaroni, University of Milan / Department of Environmental Science and Policy; C.D. Possenti, B. De Felice, Università degli Studi di Milano; N. Saino, University of Milano

Oviparas transfers mother to the eggs components that have both independent and combined effects on offspring phenotype. Functional interactions between egg components, such as antioxidants and contaminant compounds, are expected to change in the concentration of one component has effects on offspring traits. The dependent of the concentration of other interacting components. However, the combined effects of variation in different egg components are virtually unknown. Bird eggs contain vitamin E (VE), a major antioxidant, and also a variable amount of maternally-transferred contaminants. Polybrominated diphenyl ethers (PBDEs) are a family of brominated flame retardants that have been widely used as non-ionic additive compounds diverse commercial products. Many monitoring studies have revealed the presence of PBDEs in the biota, which can induce a plethora of adverse effects at different organisms' life stages, often mediated by the onset of oxidative stress. Although PBDEs have been found in birds and their eggs, the consequences of environmental contamination in several bat species. Until now, no study has considered that the oxidative stress-related toxicity of these compounds may be counteracted by the presence of antioxidant molecules that mothers allocate to their eggs at the time of laying. The independent consequences of variation in the egg concentrations of VE and PBDEs on offspring phenotype, including morphological and oxidative stress effects, are largely unknown, while no study has investigated their combined effects. Thus, we manipulated the concentration of VE and BDE-47, a PBDE congener having a well-known pro-oxidant activity, in the eggs of wild yellow-legged gull (Larus michahellis) by administering a physiological, large (2 standard deviations) dose of VE and 150 ng yolk of BDE-47 both independently and in combination. We tested for effects on morphological traits (body mass, skeletal growth) and oxidative stress, as changes in total antioxidant capacity, amount of pro-oxidant species, antioxidant enzyme activity, lipid peroxidation, protein carbonylation and DNA fragmentation, in embryos soon before the hatching.
MO087

Sensitivity of freshwater pearl mussel juveniles (Margaritifera margaritifera) to different environmental and contamination factors

Th. Belamy, University of Bordeaux; A. Legay, University of Bordeaux / UMR EPoC CNRS 5805; B. Etcheguierr, University of Bordeaux / UMR CNRS 5805 EPoC; M. Baudrinont, Université de Bordeaux / UMR EPoC CNRS 5805

Margaritifera margaritifera is a freshwater bivalve mollusk threatened with extinction in Europe. The population of this freshwater pearl mussel has declined by 90% in Europe, during the XIXth century because of pearl fishing and from the XXth century due to habitat degradation, decrease in water quality and pollution. Nowadays, french population of M. margaritifera is estimated at 100,000 individuals with the largest population found in the river Dronne (Bordeaux - FRANCE) with up to 15,000 individuals. Freshwater pearl mussels are considered as an excellent indicator of aquatic ecosystem health since they require high water quality and they filter up to 50 L of water a day. As a result, they are called «umbrella species», meaning that their conservation will benefit all species living in the same river. With the aim of preserving this pearl mussel, the European project LIFE «Preservation of Margaritifera margaritifera and restoration of river continuity of the Upper Dronne river 2014-2020 » has been set up in which a farm was created in order to produce juveniles in captivity. Some of them will be reintroduced into the environment while others will be used for ecotoxicological studies. The aim of this work was to determine the sensitivity of M. margaritifera juveniles to different environmental and contamination factors, since they are considered as the most sensitive life stage of this species. Acute toxicity tests were carried out on one-year-old juveniles in order to determine toxicity thresholds (LC50) of several factors such as temperature, dissolved oxygen, nitrates, phosphates and metals. Those data will allow to target reintroduction areas of juveniles produced in the farm and help the conservation strategies of Margaritifera margaritifera in the Upper Dronne river.

MO088

Using population modelling to reduce uncertainty - an example of a herbicide

M. Wang, WSC Scientific GmbH / Dept Etathe Modelling; M. Foudoulakis, Dow Agrosciences / RSRA ERS

Uncertainty is perceived as a major acceptability criterion for higher tier risk assessment. While uncertainty does not start to be present at the higher tier (though it is generally not considered in the first tier) it needs to be addressed explicitly when higher tier refinements are applied which make the risk assessment more realistic. The use of standard higher tier refinements or even modelling is sometimes perceived as increasing uncertainty. However, in the present poster we demonstrate that refinements and population modelling help to reduce uncertainty by providing additional insight into the mechanistic understanding of risk and the ecological relevance of effects. This is done based on a risk assessment for a herbicide. It is shown how what-if questions help to address specific questions on uncertainty in the risk assessment and how a margin of safety can be calculated.

MO089

SETAC Wildlife Toxicology Interest Group

J.E. Elliott, Environment Canada / Science Technology Branch

LCA method developments in a global perspective: Status and outlook (P)

MO090

A tool to integrate consumer and environmental exposure in life cycle impact assessment

Q. Jolliet, University of Michigan; L. Huang, University of Michigan / Dept of Environmental Health Sciences; P. Funtke, Technical University of Denmark / Quantitative Sustainability Assessment Division

Direct exposure of consumers to chemical ingredients within our daily products is an important pathway that often dominates environmental performance profiles of these consumer products, but has been currently left aside in LCA toxicity characterization. The aim of the present study is to update and extend the existing framework to consistently incorporate consumer exposure pathways in a way fully compatible with existing LCA toxicity characterization methods, and to illustrate it via a case study of plasticizer chemicals in building materials. We developed a general a framework and a tool that extends the toxicity assessment to the near-field and consumer exposure assessment and combines it consistently with the USEtox fate and environmental exposure models. The chemical mass per functional unit in the consumer product is multiplied by the product intake fraction (PiF) to yields the total exposure expressed. The PiF represents the fraction of the chemical in products that is taken in by the consumer. It is determined by coupling fate processes in consumer environments (near-field) with existing environmental compartments and processes (far-field), via a consistent and mass balance-based set of transfer fractions. The developed tool already enables to calculate characterization factors for 22 types of building products, 8 types of personal care products, 7 contact food materials and multiple cleaning product-chemical combinations. The case study of DEHP plasticizer in a vinyl flooring shows that starting from a mass of DEHP in products of 82 kg, 0.15 kg will be taken in, mostly by the household users via dust ingestion as a dominant pathway. This leads to intake doses of 0.14 mg/kg bw/day for an adult and 0.5 mg/kg bw/day for a 3 years old child. Performing a full LCA of the vinyl flooring shows that the 16% of DEHP plasticizer in flooring are associated with dominant shares of impact on human health (5%) and on aquatic environment (2%), while it is a dominant contributor to climate change impacts (59%). This case study illustrates well the importance to account for consumer exposure to chemical in product during their use. Final outcome is a consistent and quantitative framework and directly applicable tool to determine factors based on scientific consensus for assessing life cycle exposure and toxicity impacts of chemicals in LCA, as an input to the LCIA guidance efforts of the Life Cycle Initiative.

MO091

Towards the integration of an Agent-based Model into LCA framework to assess dynamic indoor air quality

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The construction sector, representing 44% of the total final energy consumption in Europe, is recognized as a major hotspot of resource use and environmental impacts. Thus, strong efforts are made to encourage the design of environmentally friendly buildings. However, the airtightness of low energy buildings has created particularly confined and polluted indoors. Indoor pollution has been raised as a major public health issue since we spend on average 80% of our time in closed spaces. Designing sustainable buildings with good indoor air quality is even more challenging since this latter is strongly influenced by occupant’s lifestyle and behavior. Life-cycle assessment (LCA) is a relevant methodology to account for impacts from indoor air while avoiding potential burden shifting from the life cycle of energy or materials used. Nevertheless, the current use of LCA faces scientific obstacles such as: (i) the inclusion of the dynamical effects of indoor pollution on human health and (ii) the consideration of the behavior of the occupants. In order to address these concerns, a model of autonomous agent has been developed structured around (i) an agent-based model Li-BIM (Live in BIM) which exerts the Phe represent human behavior, (ii) a physical model to capture the building thermal behavior, (iii) the numerical representation of the building (BIM) and (iv) an innovative indoor air quality model Be-BIM (Breathe in BIM). Li-BIM is an operational model which simulates the behavior of the occupants based on an evolved occupational cognitive and social framework. Be-BIM is currently being developed as a dynamic and localized fate model sensitive to users’ behavior and the environmental fate of volatile organic compounds. Therefore, Be-BIM will: (i) generate the inventory data for dynamic pollutant emissions and (ii) assess the local impacts from air emissions. Expected outcomes of our integrated model include characterization factors for human toxicity due to indoor air which are dynamic and spatially differentiated at the scale of the building. Eventually, our model will allow the comparison of life cycle impacts of different building scenarios with a specific focus on indoor air quality suited for residential dwellings.

MO093

Adding the resource dimension to the WULCA framework on assessing freshwater use in LCA

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Effective management of freshwater resources is recognized as being vital. At present, existing LCA methods for water use do not entirely reflect the state of such a vital resource remaining for future generations. Thus, the objectives of this research are: (1) identify how freshwater resources can be defined as an entity to protect within the Area of Protection (AoP) natural resources, (2) describe the impact pathways affecting this resource, and (3) propose a characterization framework to assess the impacts from the identified impact pathways. Freshwater resource has a particular status in LCA resource modeling. First, it exists in the form of three types of resources: flow, fund, or stock. Then, in addition to being a resource for human economic activities (e.g. hydropower), it is above all a non-substitutable support for life that can be affected by both consumption (source function) and pollution (sink function). Therefore, both types of elementary flows (emissions and water consumption) should be linked to a damage indicator for
freshwater as a resource. In order to clearly define what is to be protected, the freshwater resource is put in perspective through the lens of three safeguard subjects. Considering the current scope of the AoP natural resources, the complex nature of freshwater resources and the dimension of freshwater to safeguard, a definition of freshwater resource is proposed. Also, a wide range of possible impact pathways to freshwater resources is identified, establishing the link between different sources and the potential to cause freshwater depletion or pollution in the long-term. The concept of recovery period is used to operationalize this framework: when the recovery period lasts longer than a given period of time, impacts are considered as being irreversible and fall into the concern of freshwater resources protection (i.e. affecting future generations).

The study shows that it seems relevant to include this concept in the impact assessment stage in order to discriminate the long-term from the short-term impacts, as some dynamic fate models already do. Recommendations are also given for freshwater resource impact indicator(s). Therefore, such an indicator would allow LCIA to capture potential long-term impacts that could transparently advise decision makers about potential safe water supply issues in the future.

**MO094**

**Considering water and soil conservation works in Life Cycle Assessment: focus on contour ridges and erosion impacts**

M. Jouini, Montpellier SupAgro / Département de génie rural; R. Campanili, IRD, UMR LISAH; S. Follain, Montpellier SupAgro, UMR LISAH; J. Burte, CIRAD / UMR GEAN; N. Benaisa, National Agronomic Institute of Tunisia / Science de la production végétale; C. Sinfort, ITAP, Irstea, Montpellier SupAgro / Département de génie rural; R. Ciampolillo, University of Michigan. The purpose of this study is to intergrate the impact of contour ridges on soil quality in LCA. There are different types of WSCW with different functions and they act differently on erosion process. In this study we focussed on contour ridges as a type of conservation works because they are associated to crop systems. Contour ridges are generally built in upland areas to reduce runoff and erosion, to increase on-site deposition of eroded particles and to increase local water infiltration. Contour ridges modify water and soil flows at catchment scale, so it is necessary to use a model able to calculate the inventory flow at the catchment and not only at the plot level. In this study we present a methodology to integrate the impact of contour ridges on topsoil erosion at the catchment level and to compute characterization factors in presence of such WSCW. The proposed method was applied in a case study in semi-arid context in central Tunisia (Merguellil watershed) which presents the issues of over-exploitation of water resources, accelerated land degradation and a high expansion of conservation works. In order to highlight the impact of WSCW on soil quality in LCA, different methods and techniques were tested. For life cycle impact assessment, we focussed on two midpoint impact categories on soil quality of LANCA model: erosion resistance and mechanical filtration. The results showed how contour ridges can modify topsoil erosion process and therefore the impact on soil ecological functions for several production systems. In conclusion, it is necessary to integrate the positive impacts of WSCW during the whole life cycle assessment. However, it is not possible to compute these impacts because they are associated to crop systems.

**MO095**

**Impact of heavy metals on human toxicity using LCA: a case study for Wallonia**

S. Gerbinet, Université de Liège / Chemical Engineering; F. Van Stappen, CRAW Walloon Agricultural Research Centre; S. Belboom, ULiege; E. Pezennec, Knauf Insulation spr.; S. Groselambert, University of Liège - Chemical Engineering / Dpt of Chemical Engineering - PEPS; A. Leonard, University of Liège

This study aims on summarising results when assessing the human toxicity of corn farming in Wallonia, Belgium. The USEtotx method is applied to the farming of one hectare of corn. Local data are used for farming data and GaBi datasets are used for background data. The field emissions due to farming are calculated by the most used models. The results in human toxicity, cancer effect, underlie the large contribution of chromium (Cr) emissions due to the use of organic and mineral fertilisers. But despite the high contribution of chromium emissions, it is, in USEtotx, the average of the one of Cr (III) (non-toxic) and Cr (VI) (toxic), therefore really larger than the one of Cr (III). Therefore, a test is realized where 95% of the chromium is present as Cr (VI) and the rest is Cr (III). In this case, score in human toxicity cancer effect is divided by 7, whereas this has no influence on the other results. The impact for human toxicity, non-cancer effect is mostly related to zinc emissions in soil due to the use of organic fertilizers.

**MO099**

**Combining use of Mixed-Integer Optimisation and Thermodynamic, Molecular and Charge Density attributes for predicting Life Cycle Production**

S. Groslambert, University of Liège - Chemical Engineering / Dpt of Chemical Engineering - PEPS; A. Leonard, University of Liège

The purpose of this study is to intergrate the impact of contour ridges on soil quality in LCA. There are different types of WSCW with different functions and they act differently on erosion process. In this study we focussed on contour ridges as a type of conservation works because they are associated to crop systems. Contour ridges are generally built in upland areas to reduce runoff and erosion, to increase on-site deposition of eroded particles and to increase local water infiltration. Contour ridges modify water and soil flows at catchment scale, so it is necessary to use a model able to calculate the inventory flow at the catchment and not only at the plot level. In this study we present a methodology to integrate the impact of contour ridges on topsoil erosion at the catchment level and to compute characterization factors in presence of such WSCW. The proposed method was applied in a case study in semi-arid context in central Tunisia (Merguellil watershed) which presents the issues of over-exploitation of water resources, accelerated land degradation and a high expansion of conservation works. In order to highlight the impact of WSCW on soil quality in LCA, different methods and techniques were tested. For life cycle impact assessment, we focussed on two midpoint impact categories on soil quality of LANCA model: erosion resistance and mechanical filtration. The results showed how contour ridges can modify topsoil erosion process and therefore the impact on soil ecological functions for several production systems. In conclusion, it is necessary to integrate the positive impacts of WSCW during the whole life cycle assessment. However, it is not possible to compute these impacts because they are associated to crop systems.

Therefore, the US EPA has developed a research project to improve such characterization. Several recent papers have suggested that the exposures to these consumer products and building materials may be significantly greater than exposures to far-field emissions, and therefore, not including these exposures may result in decisions which are unknowingly biased in a manner which could lead to increased risk. The difficulty up to this point in time has been to characterize these exposures since the product compositions are often unknown and the pathways to exposure to these products is becoming recognized as increasingly important. Therefore, the US EPA has developed a research project to improve such characterization.

**MO099**

**Comparing ProScale Hazard Factors with USEtox Effect Factors for human toxicity**

T. Rydberg, IVL Swedish Environmental Research Institute; H. Holmquist, Chalmers University of Technology. The purpose of this study is to compare side-by-side, the Hazard Factors (HF) of ProScaleTM, and the Effect Factors (EF) for human toxicity of USEtoxTM, and analyse the results, as both factors have been developed as a metric for adverse human health impacts due to toxic effects. Hazard factors in ProScale are derived based on substances classification in the GHS/CLP classification system, reflecting health effect severity based on Hazard class and a dose-response based on Solving Grouping in five ProScale hazard classes, and an OEL based correction factor has been introduced o account for potency within each class. The effect factor (EF) is a metric of the change in life time disease probability due to change in life time intake of a pollutant (cases/kg). USEtoxTM determines effect factors for carcinogenic and noncarcinogenic chemicals separately. Both methods have separate factors for inhalation and oral exposure routes. However, with increased use of LCA to support decisions related to chemical alternative assessments, characterization of the near-field exposures to these products is becoming recognized as increasingly important. The views expressed in this abstract are those of the authors and do not necessarily represent the views or policies of the U.S. Environmental Protection Agency. Mention of trade names or commercial products does not constitute endorsement or recommendation for use.
Impacts of Chemicals
R. Calvo-Serrano, G. Guillén Gósáulez, Imperial College London / Chemical Engineering

Process sustainability has become one of the fundamental criteria for decision making in chemical industry, being Life Cycle Assessment (LCA) the most popular method in recent years, currently being one of the most extended sustainability assessment methods. Since LCA is based on the analysis of all interactions for all the stages of the life cycle, it ends requiring large amounts of information. This information, however, can be difficult or impossible to gather, being one of the main obstacles when trying to apply LCA. Chemical industry is particularly affected, easily having thousands of interactions even for small and relatively simple processes and only information of a few hundreds. In these cases, when a full LCA cannot be applied, a simplified version is used instead. These Streamlined LCA (SLCA) follow the same basis as LCA, but generally either simplify the scope of the analysis and/or reduce the amount of information required in the assessment.

The precise simplifications to be done (and the assessment discrepancy with the full LCA) have to be specifically considered for the process or activity assessed. Under these principles, we present a novel approach for the estimation of LCA impact categories associated to the production of chemicals using information of their chemical and physical properties. We propose that the physical properties of the products are directly related with the impacts generated in the production process, and that these impacts heavily contribute to the overall impact generated for the production of the chemical analysed. Previous studies demonstrated the prediction capabilities of molecular and thermodynamic attributes. Here we also consider the ρ-effector model as attributes, for a better characterization of the chemicals and therefore to the generated impacts. In addition, we propose to reduce the whole assessment methodology to linear prediction models, selecting in each model only the attributes that better describe specific impact categories. This approach has been applied to a database consisting of 83 chemicals, considering 15 molecular descriptors, 17 thermodynamic attributes and 8 ρ-profile partitions, being able to obtain reasonable LCA impact estimates such as Global Warming Potential (GWP)3.35% or Eco-Indicator09 (EIP)18.34%.

MO100 Development of USEtox characterisation factors for micropollutants in effluents
E. Maillard, ELSA-PACT Industrial Chair

Many substances are increasingly detected in surface waters, after their use by the human population. In most cases, these substances will exert the same effects as desired when they are originally applied, only now affecting different organisms. These effects can occur at concentrations of µg/l, which is why these substances are called micropollutants. In the context of Life Cycle Assessment, there is a need of characterising the toxicity potential of these micropollutants affecting ecosystems and/or the human population. A substance which is not characterised will not be considered in a LCA study, which may result in misguided decisions and the omission of essential environmental issues related to biodiversity and human health. The aim of this project is to develop a database of characterisation factors for the most micropollutants as attributes, for a better characterisation of the substances and therefore to the generated impacts. In addition, we propose to reduce the whole assessment methodology to linear prediction models, selecting in each model only the attributes that better describe specific impact categories. This approach has been applied to a database consisting of 83 chemicals, considering 15 molecular descriptors, 17 thermodynamic attributes and 8 ρ-profile partitions, being able to obtain reasonable LCA impact estimates such as Global Warming Potential (GWP)3.35% or Eco-Indicator09 (EIP)18.34%.

MO101 Adjustment of freshwater ecotoxicity with USEtox
M. LOT, CEHTRA; P. Thomas, CEHTRA SAS; F. Sahigara, KREATIS; M. Jacob, TOTAL SA

USEtox is determined as the reference tool for freshwater ecotoxicity impact evaluation in LCA context. By the way, it is recommended by several institutes: by European Commission for PEFOEFOE project, by JRC-IES in ILC framework, by WHO in their Guidelines on chemical and by US- EPA in TRACI tool manual. An assessment of the relevance of the Characterisation Factor (CF) with its associated factors was realized. We found that CF appears to be significantly influenced by the effect factor (EF), implying that the 2 others factors, XF (Exposure Factor) and FF (Fate Factor), do not intervene, or very little, in the final calculation of CF. This finding is surprising because the XF and FF factors should influence the calculation of CF as they represent, significant fate adjustments: substance bioavailability (XF) and its presence in the medium (FF). Hence, the influence of each factor was analysed in more detail and some inconsistencies were noted. For EF, USEtox includes an extrapolation calculation for acute to chronic toxicity (called Acute-to-Chronic Ratio). This extrapolation is not always reliable and will certainly not be applicable for substances with a log Kow<6 where acute toxicity is superior to solubility but chronic toxicity may still occur. Moreover, the value used for the ACR seems inappropriately low and should be different according to the mode of action of the substance. For XF, adsorption starts to reduce XF at log Koc around 5 whereas adsorption of organic substances is generally considered to become highly significant in ecotoxicological studies performed at low concentrations from log Koc of 4. Overall, the XF seems to be overestimated in this model for the majority of substances with a log Kow between 3 and 5. For FF, one of the parameters that most influences its calculation seems to be the biodegradation, which alone can alter the relationship between the EF and the CF by an order of magnitude (between a highly persistent and a highly biodegradable compound). However, the relative influence of biodegradation (and the other FF parameters) on the CF is too limited compared to the EF value. Indeed, the difference between the minimum and maximum biodegradation on CF is approximately 2 orders of magnitude while the EF itself spans at least 8 log units. Thus, because of these inconsistencies, it is crucial to discuss the relevance of each factor with the aim of improving the model providing a more realistic approach.

MO102 Advancing nutrient modelling in eutrophication methods for life cycle impact assessment

LCA is better informed by scientific decisions that affect water quality. Anthropic contributions of the nutrients nitrogen (N) and phosphorus (P) threaten the health of freshwater and marine ecosystems around the world [1]. The increase in environmental nutrient availability, known as eutrophication, can lead to harmful algal blooms (HABs) and decreased levels of oxygen (hypoxia) needed to sustain aquatic life [2]. This presentation evaluates the current state of life cycle impact assessment (LCA) methods for eutrophication in freshwater and marine ecosystems using a criteria-matrixed review of the underlying fate and transport (F&T) models. Results of the review point to several key recommendations for further scientific development of mid-point eutrophication methods in life cycle assessment (LCA). Current eutrophication methods in LCA are limited. Most LCA studies use simplified F&T models with continental or global geospatial resolution, characteristics of which tend to be inadequate for regional analyses and complex community-based decisions. Therefore, this critical review examines a set of surface water quality models, watershed models, marine models, and air quality models that each have potential for integration into LCIA. Factors examined include sources of nutrient loading to each environmental compartment (e.g., water, agriculture, energy production, etc.), the forms of each nutrient modeled, and the representation of each F&T mechanism. Review results suggest several possible recommendations, including the continuation of the recent trend toward separation of freshwater and marine eutrophication methods, expanded characterization of the freshwater cause-effect chain, and the development of new soil and freshwater fate factors. By incorporating findings of the F&T models into current eutrophication methods, LCA can better inform scientific decisions that affect water quality, nutrient management, and environmental policies across watersheds and global ecosystems. [1] Rockström J, Steffen W, Noone K, Persson Å, Chapin FS, Lambin EF, Lenton TM, Scheffer M, et al. 2009. A safe operating space for humanity. Nature 461: 472-475. [2] Diaz RJ, Rosenberg R. 2008. Spreading dead zones and consequences for marine ecosystems. Science 321: 926-929. Disclaimer - The views and policies of the U.S. Environmental Protection Agency do not necessarily represent the views or policies of the U.S. Environmental Protection Agency / National Risk Management Research Laboratory. Mention of trade names or commercial products does not constitute endorsement or recommendation for use.

MO103 Land Use Change comprehensive framework in LCA for microbial cultivation systems as emerging production option in the bio-economy
L. Compagnoni, D. Manzana, University of Bologna; S. Righi, University of Bologna / Physics; E. Balugani, E. Merloni, University of Bologna / Physics; E. Balugani, E. Merloni, University of Bologna

Europe is nowadays facing serious issues about natural resources depletion. Post-industrial societies will enhance the transition from a fossil- based economy to a bio-based society. In this context, algae represent an emerging resource of great importance for their potential applications. Specifically, microalgae are currently promoted not only as fuel-sources, which have been studied since decades, but also as high-value products useful in pharmacy, cosmetics, nutra-tecues as well as for aqua- and agricultural uses, hence considered as high added value products. The bio-economy concept will enhance several LCA of algae-based products have been performed on a wide range of production processes. A Scopus review on “algae LCA”, indeed, reported 228 total papers published in the scientific literature since 1989, experiencing a fast-growing trend from 2010 onwards, mostly regarding biodiesel (>77%). However, one impact category of the algal-based product life cycle that is commonly overlooked, while being of high importance for the bio-economy, regards land use change (LUC) with only 8% of algae’s LCA-related studies including it. Land use influences biodiversity as well as the structure and functions of ecosystems, causing damage to many areas of protection through diverse impact pathways, such as Biodiversity.
Damage Potential, Ecosystem Services Damage Potential, Biotic Natural Resource Depletion and Climate Change. However, only few of these impact pathways are fully implemented in currently available LCIA tools, also due to lack of significant consensus on this novel impact category. Specifically, LUC has to be carefully evaluated when assessing microalgae’s cultivation systems, as they may be strongly diverse one each other, hence impacting through diverse paths. Cultivation layouts may range, in fact, from large open ponds to more compact photobioreactors; they may be installed in natural environments such as fresh water ponds or offshore cultivation systems, either in brownfield lands in an optic of redevelopment of industrial areas, hence even generating a positive effect to the environment, mostly in terms of GHG’s fluxes and biodiversity. In this respect, the study aims at providing a consistent framework of the current methodology on LUC impact category and its application to bio-economy and, specifically, to microalgae’s production in order to provide support to business and policy decision making.

MO104 Application of LCIA water use methods to renewable energy systems in Spain
L. Sánchez-De Castro, D. Garrain, Y. Léchón, CIMAT / Energy Dpt Energy Systems Analysis Unit

The topic of ‘water’ in LCA has emerged as an important approach to quantify the related effects of water use from consumption of goods and services. Several assessment methods have been proposed by the scientific community, encompassing both the computation of water use and its impacts, but differing in the run off or recharge in the ground. After decades of continuous evolution of the methodologies to consider this impact category, this work presents the application of the most relevant methods for quantifying the water use in LCA of several renewable energy systems in Spain: i) Water Footprint standard harmonized by means of ISO 14046, and ii) the AWARE method developed by WULCA working group (http://www.wulca-waterlca.org), a midpoint water use indicator representing the relative Available Water Remaining per area in a watershed.

MO105 Identification of methodological challenges remaining in the assessment of a water scarcity footprint
P. Mariotto, University of Aveiro / Department of Environment and Planning; b. Ridoutt, CSIRO; L. Arroja, A. Dias, University of Aveiro / Department of Environment and Planning

Freshwater stress and its implications for present and human welfare and the natural environment awakened the need to develop spatially explicit methods to calculate the product water footprint (WF) from a life cycle perspective. In this sense two parallel developments emerged: a water footprint assessment (WFA) developed by the Water Footprint Network (WFN) to map direct and indirect water use along supply chains and its relevance in water resources management; and an impact-based WF following the life cycle assessment (LCA) methodology, according to the ISO 14046. Both WFN and LCA-oriented impact assessment methods considers blue water (fresh surface and groundwater) and green water (rainfall on land that does not run off or recharge the groundwater). After decades of continuous evolution of the methodologies to consider this impact category, this work presents the application of the most relevant methods for quantifying the water use in LCA of several renewable energy systems in Spain: i) Water Footprint standard harmonized by means of ISO 14046, and ii) the AWARE method developed by WULCA working group (http://www.wulca-waterlca.org), a midpoint water use indicator representing the relative Available Water Remaining per area in a watershed.

MO106 Filling the Gap of Overfishing in LCIA: Eco-factors for Global Fish Resources
M. Sticku, Zurich University of Applied Sciences / Institute of Natural Resource Sciences; L. Eymann, ETHZ Swiss Federal Institute of Technology; B. Keesler, R. Iten, Zurich University of Applied Sciences / Institute of Natural Resource Sciences

There is a lack of LCIA methods to assess the contribution of fish consumption to the global problem of overfishing. Due to this methodological gap, fish is often determined to have a lower environmental footprint than other sources of animal protein in LCA studies. The aim of this publication is to present the development of eco-factors for fish resources and by-catch as an addition to the Swiss Ecological Scarcity Method 2013 and to compare the eco-factors of different fish resources and the impacts of fishing as well as the impacts of dietary alternatives like chicken, pork, lamb, beef and veal. The sustainable use of fish resources ensures that the fish stock for each fish species exceeds the minimum stock needed for a maximum sustainable yield. Therefore, the method developed uses a distance to target approach based on the fish stock for maximum sustainable yield (FSMSY) and the current fish stock (FS) for the characterisation of the use of fish resources. The eco-factors were calculated for each fish species and fishing area. The inclusion of the eco-factors shows the relevant contribution to the total environmental impact of fish compared to fishing and fish processing. Depending on the fish species, fish can have significantly higher overall impacts compared to different types of meat. The contribution of the eco-factors for by-catch is minor compared to fish resources but still relevant for the comparison with dietary alternatives. Distance to target based eco-factors using regionalised data for FS and FSMSY not only follow a comparable underlying approach as suggested in literature but also enable aggregation into a single-score with other environmental impacts. The overexploitation of fish resources is highly variable for different fish species and fishing areas as well as for by-catch. When comparing fish to equivalent dietary alternatives like chicken, pork, lamb, beef or veal, consideration of the overexploitation of fish resources results in some fish species exceeding the environmental impact of dietary alternatives. Therefore, the overexploitation of fish resources is relevant in the Life Cycle Assessment of fish products in different diets. The stated approach can be applied for each of the three categories of LCIA of meals and is able to reflect the regionalised impacts caused by the overexploitation of fish resources.
freshwater eutrophication due to nitrogen as well as phosphorus emissions from wastewater treatment plants (WWTPs) and agriculture on the global scale. CFs were defined as the change in not potentially occurring fraction of species (PNOF) due to a change in the river basin-specific emission of P or N to freshwater (via WWTPs) or agricultural soil and consist of a fate and an effect factor. To determine the fate factors, the change in N and P concentrations resulting from agricultural as well as from WWTPs from year 2000 compared to year 1900 were separately modeled for every river basin in the world. Effect factors were based on log–logistic relationships between the PNOF (dimensionless) of heterotrophic species and total P (TP) or NO3 concentrations. The PNOF – concentration relationships were determined using data on the highest concentration where a species was observed in field surveys. Our work provides the opportunity to quantify worldwide spatially-explicit and cumulative impacts due to nitrogen as well as phosphorus from soil as well as from freshwater emissions, in a coherent way. The factors can be applied to determine eutrophication impacts of products in LCIA, as well as to determine country-specific eutrophication footprints using multi-regional input-output (MROI) analysis.

Building of large-scale inventories of emissions and resources and applications for environmental footprints of territories, nations and sectors (P)

MO109 Carbon and material footprint of consumption in Flanders - an input-output based assessment
A. Vander Putten, VITO NV; K. Boonen, M. Christis, VITO; E. Vander Putten, VMM-MIRA
Environmental footprints of a country or region are a measure for the environmental impact that is caused worldwide by national or regional consumption. The Flemish Environment Agency (VMM) asked VITO to assess the carbon and material footprint of consumption in Flanders (region of Belgium) and identify hot spots and bottlenecks, based on the Flemish environmentally extended input-output model. The Flemish EE-IO model is based on specific monetary and environmental data for the Flemish region of Belgium and is part of an interregional IO-model in which trade with the Brussels and Walloon Region is modelled in IO-tables. Imports from outside Belgium are included via the link with the use table of Ecoinvent. A monetary concentration matrix for households divides the final demand vector of households in different consumption categories. In 2010 the carbon footprint of Flanders amounted to about 20 tonnes per inhabitant. Nearly three quarters of the carbon footprint are linked to household consumption, mainly caused by the production and transport of the goods and services consumed. Three quarters of the carbon footprint of goods and services purchased by households are linked to housing, food and personal transport. Whilst the majority of the greenhouse gas emissions, primary materials and employment is outsourced, the added value linked to Flemish consumption is mainly created in Flanders. The presentation will introduce the overall results of the carbon and material footprint assessment of Flemish consumption in 2010 and go more in detail into the value chain impact of some household consumption activities e.g. food consumption. The relation between carbon and material footprint, geographical and sectoral distribution of different production chains and consumption activities, and the relation with added value and employment created by Flemish consumption will be discussed. As the model is available for 2003, 2007 and 2010, the evolution over these years will also be presented. Conclusions will mainly focus on methodological issues and policy implications that follow from this analysis. To achieve the greatest possible global environmental benefit, it is not enough to focus on a country or region’s boundaries alone. There is also a need for a policy that is aimed at making production chains and consumer behaviour more sustainable, including internationally harmonised calculation methods and targets of footprints to evaluate the results.

MO110 A cross-country analysis of relationship between economic structural change and CO2 emissions
K. Shironitt, Kyushu University / Economics; S. Kagawa, Kyushu University; Y. Kondo, Waseda University
In this study, we focus on the effects of changes in material and energy input structure on the life-cycle CO2 emissions (i.e., consumption-based emissions). Previous studies have conceptualized the economic structural change including the shift toward a service economy and the increase in greenhouse gas (GHG) emissions embodied in consumptions of a specific country (United States or Japan) (Suh, 2006; Nansai et al., 2009). This study is an important follow-up research that examines the environmental effects across countries and evaluates whether or not the development levels of countries can explain these environmental effects. Specifically, we employed a multiple logistic structural decomposition analysis based on the World Input-Output Database (WIOD) during 1995 to 2008 (Dietzenbacher et al., 2013) and decomposed life-cycle CO2 emissions of 40 nations into the following four inducement sources: (i) inputs from material goods (including energy) to material goods, (ii) inputs from material goods to services, (iii) inputs from services to material goods, and (iv) inputs from services to services. From the results, we found that the role of inputs from material goods to services and inputs from services to material goods in increasing life-cycle CO2 emissions have become more important than inputs from material goods to material goods in not only developed countries but also developing countries. Services are sustained by manufactured goods, so manufactured goods are necessary for the continued growth of service sectors. In a developed country, a large proportion of its GDP is generated by tertiary sector activities, and the role of service sectors in economic growth will expand more and more. In developing countries too, since the proportion of production from primary and secondary industries will shrink and that from tertiary industries will rise with their economic growth, the importance of service sectors will be enlarged. For this reason, it is essential to focus on the production systems of service sectors to reduce domestic CO2 emissions. It is especially important to sift to the industrial structure with less emission-intensive material goods.

MO111 Influence of substance coverage on impacts from the electricity sector
A.S. Leclere, DTU / Management Engineering; M.Z. Hauschild, Technical University of Denmark / DTU Management Engineering – Division for Quantitative Sustainability Assessment; R. Wood, Norwegian University of Science and Technology / Biology; A. Laurent, DTU / Division for Quantitative Sustainability Assessment DTU Management Engineering
The electricity sector is a major source of emissions of greenhouse gas, but also heavy metals, dioxins or radioactive isotopes. However, most environmental assessments of the electricity sector at national or global scale focus solely on climate change and do not include other environmental impact categories such as particulate matter formation or toxic impacts on human health. At the national scale, the few available databases are limited to a narrow substance coverage. For example, official reports of pollutants emissions to the European Monitoring and Evaluation Programme (EMEP) should cover 23 substances in 51 countries, but they are not always complete. The Multi-Regional Input-Output database E2010-SE includes emissions to air for at least 44 countries and 5 regions but only for 33 substances. In comparison, the database Ecoinvent provides emission data for hundreds of substances in the unit process inventories for electricity and heat generation. Here, we aim to develop a globally consistent and extensive dataset of airborne emissions from electricity production to get a more realistic coverage of toxicity impacts in large-scale life cycle assessments (LCAs). We thus built the Ecoinvent-based National Energy-related Emission Inventory (EENEE) by upscaling processes from Ecoinvent 3.3 with national production volumes of electricity and complementing it with emission data from external sources. The resulting inventory EENEE covers 229 substances, including 51 radioactive isotopes. By comparing inventories and databases at midpoint level, we show that using Ecoinvent may underestimate the toxicity impacts associated with electricity production by a factor ranging from 1.4 to 1.9, while Ecoinvent may cut them off by up to 4 orders of magnitude in some countries. This demonstrates the importance of having an extensive substance coverage to fully represent the environmental impacts of electricity production.

MO113 Mapping the carbon, air pollution, and biodiversity footprints of nations: A GIS + global supply chains approach
K. Kanae, Shinsyu University / Faculty of Economics and Law; D. Moran, Norwegian University of Science and Technology
"Spatial footprinting" is an approach for locating the actual hotspots where impacts due to consumption occur. As a result, using footprinting offers the potential to link any remote sensing or earth observatory GIS data that is tagged to an economic sector to spatial footprinting offers the potential to link any remote sensing or earth observatory GIS data that is tagged to an economic sector to spatial footprinting offers the potential to link any remote sensing or earth observatory GIS data that is tagged to an economic sector to spatial footprinting offers the potential to link any remote sensing or earth observatory GIS data that is tagged to an economic sector to spatial footprinting offers the potential to link any remote sensing or earth observatory GIS data that is tagged to an economic sector to national or international footprinting. For example, official reports of pollutants emissions to the European Monitoring and Evaluation Programme (EMEP) should cover 23 substances in 51 countries, but they are not always complete. The Multi-Regional Input-Output database E2010-SE includes emissions to air for at least 44 countries and 5 regions but only for 33 substances. In comparison, the database Ecoinvent provides emission data for hundreds of substances in the unit process inventories for electricity and heat generation. Here, we aim to develop a globally consistent and extensive dataset of airborne emissions from electricity production to get a more realistic coverage of toxicity impacts in large-scale life cycle assessments (LCAs). We thus built the Ecoinvent-based National Energy-related Emission Inventory (EENEE) by upscaling processes from Ecoinvent 3.3 with national production volumes of electricity and complementing it with emission data from external sources. The resulting inventory EENEE covers 229 substances, including 51 radioactive isotopes. By comparing inventories and databases at midpoint level, we show that using Ecoinvent may underestimate the toxicity impacts associated with electricity production by a factor ranging from 1.4 to 1.9, while Ecoinvent may cut them off by up to 4 orders of magnitude in some countries. This demonstrates the importance of having an extensive substance coverage to fully represent the environmental impacts of electricity production.

MO114 LCA data machine applied
A. Ciroth, GreenDelta; M. Stroeka, GreenDelta GmbH
In any LCA study, finding data sets that are “fit for purpose” is probably one of the aspects which consumes most time and effort, and the data sets which are used have obviously a direct implication on the stability of the result achieved. This high effort for finding and creating data sets is true despite the existence of several comprehensive, generic databases for LCA. It is underlined by the presence of intraclass, outdated, or out-of-region and context data set in these databases. For this reason, an “LCA data machine” has been developed at GreenDelta in the last 3 years, and was already presented earlier at conferences. The LCA data machine automatizes creation, update, and to some extent also review of data sets in LCA and sustainability assessment. Data sets are created to meet several specific requirements, e.g. related to region, time, or nomenclature system, but can also be

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created so that they meet requirement sets, such as, for example, related to PEF.

Meanwhile, the “DAMA” has been applied to various sectors, products, and data sources. The presentation will summarise key steps of the development and will demonstrate the DAMA, for specific data sets and also for specific use cases. The LCA data machine will be demonstrated in three different application cases: 1) finding and if necessary creating data sets in situations where no data set is directly available, i.e. for data gaps; Paper machine example 2) creating a data set as copy of an existing process, to adapt to specific, local needs; Creating soy bean production US 3) Product comparison, identification of differences between compared products The approach with DAMA will also be compared to approaches currently used in LCA, with examples from the PEF remodeling project and others. The LCA data machine has the potential to truly change the current approach for data set creation, exchange, and also use, in LCA and related areas and can be especially useful for creating inventories in a larger scale.

MOI15 Static and dynamic modeling of high performance buildings: Comparison of alternative scenarios in the electricity mix, a consequential effect on LCA results M. Bilec, W. Collinge, University of Pittsburgh / Civil and Environmental Engineering; H. Rickenbacker, University of Pittsburgh / Civil & Environmental Engineering; A. Landsis, Clemson University / Environmental Engineering and Earth Sciences; C. Thiel, New York University School of Medicine / School of Population and Public Health

Tantalising one's green orientation (LCA) involves explicit assumptions and major uncertainties associated with the source of electricity across two main dimensions: spatial and temporal. There is a need within the LCA community for an approach that addresses this ambiguity and allocates environmental impacts as a function of marginal and time-specific variabilities. In our study, particular attention is paid to the dynamic characteristics of two buildings’ electrical energy consumption in relation to the power grid. We evaluate two scenarios: One LCA model incorporates hourly energy use data for on-site renewable production at a net-zero energy building (NZEB) and hourly sub-hourly electrical energy usage data at a LEED Gold building; both are situated in an energy conservation district located in Pittsburgh, PA. Seven iterations of both static and dynamic life cycle assessment (DLCA) based-models were performed and evaluated based on building energy use (predicted vs. observed), electrical grid time resolution (yearly, monthly, hourly), and region-specific electrical grids and data sources (Environmental Protection Agency, Department of Energy). Our results illustrate that the use of photovoltaics at the NZEB produced excess electricity by on-site renewables which is distributed back to the grid and can be interpreted as avoided upstream emissions (generation at the power plant), which in some cases may offset or erode the aggregate material phase impacts. The marginal consequential model improved the payback period by an order of magnitude (12.5 years to 3.0 years). Additionally, the dynamic scenarios explored in this study were able to effectively account for the growth in natural gas generation, assigning or ignoring emissions based on a marginal increase or decrease load during the building’s energy use. The LEED Gold building is solely reliant on the regional electricity grid, making its findings indicative of a market-as-usual scenario and therefore comparable and/or scalable to other building studies. No studies combining time-resolved building electrical usage with time-resolved grid electricity production have been found in a U.S. context. There are notable differences between the European and North American power grids, on that account this approach aids to the advancement of DLCA research domestically.

MOI16 Life cycle framework for environmental assessment of public transport systems A. Shinde, Indian Institute of Technology Bombay, Mumbai, India; A. Dikshit, Indian Institute of Technology Bombay, Mumbai, India / Centre for Environment Science and Engineering (CESE); R. Singh, Thinkstep Sustainability Solutions Pvt. Ltd., Mumbai, India

Several studies have assessed life cycle environmental impact of public transport systems. However, there is no single platform, software or tool for comparing the environmental performance of different urban commuting processes. The objective of this study was the development of an LCA based framework to evaluate, analyze and compare the life cycle environmental impact of public transport systems. The modular and parameterized system of GaBi 6.5 platform has been used for exploring the life cycle environmental performance of commuting trains, public bus transport and intermediate public transport (IPT) modes viz. taxi, auto-rickshaw. The system boundary comprises environmental life cycle inventory, life cycle impact assessment and maintenance of transport infrastructure, manufacturing and maintenance of the vehicle, energy/fuel production and tail-pipe emissions. This framework is capable to characterize the per passenger km travelled (PKT) and vehicle km travelled (VKT) environmental impacts of an existing and proposed project. Since per PKT environmental impact of public transport systems are highly sensitive to ridership levels, the environmental impact of public transport systems and IPT modes has been compared for off-peak, average and peak hour levels of ridership. This framework has been developed on the basis of extensive data collected for the material and energy required for the construction and maintenance of infrastructure, manufacturing and maintenance of rolling stock, use phase impacts of fuel/energy production and tail-pipe emissions, in addition to the transport of raw materials.

Therefore, this framework is highly comprehensive but it is also amenable to future additions and expansions. The regional transport authorities can proactively address the target areas for improving the environmental performance of their transportation system, and ultimately the competitiveness of their network. The regulatory authorities will have the information to improve on their policies to reduce environmental impacts associated with each mode at each stage.

MOI17 Environmental impact assessment of rail freight intermodality in Belgium using the Life Cycle Assessment methodology A. L. MERCHAN, University of Liege / Chemical Engineering, PEPs; S. Groslandeb, University of Liege / Chemical Engineering; A. Léonard, Liège Université / Chemical Engineering - PEPs

BRAIN-TRAiNS is a project supported by the Belgian Federal Government that deals with the possible development of rail freight transport in Belgium, analysing the current situation of the intermodal rail freight transport from an interdisciplinary perspective. The objective of increasing the rail freight transport is linked to the European Commission’s White Paper on transport (2011), which aims to shift the 30% of road freight over 300 km to other modes of transport more energy-efficient such as rail or waterborne transport by 2030. In the framework of the BRAIN-TRAiNS project, the Life Cycle Assessment (LCA) methodology has been chosen to analyse the environmental impact of the intermodal rail freight transport in Belgium. In a first stage we have carried out the LCA of rail freight transport (distinguishing between electric and diesel traction), inland waterways transport and road freight transport independently. In a second stage we have carried out a study of the environmental impacts related to intermodal rail freight transport. For this, we have studied several consolidated intermodal rail-road routes in Belgium. The aim of this analysis is to compare the environmental impacts of these new intermodal routes depending on the freight transport mode chosen (rail or road transport) for the major part of the intermodal route. Finally, we have analysed how the increase of rail freight transport in the modal split as a result of the possible development of the intermodal rail freight transport affects the environmental impacts of inland freight transport in Belgium. For this, three divergent Belgian scenarios with a time frame set in the year 2030 have been built for further analysis. These scenarios are directly linked to the third strategic goal of the European Commission’s White Paper on transport (2011). As a result, a best, worst and medium case scenarios have been developed, depending on whether the 30% shift will have been successfully accomplished, the status quo will have been maintained or the goal will not have been completely reached by 2030, respectively. The results of this research show that the initial aim of building new intermodal rail routes in order to define the sustainability impact of future intermodal transport. They could help in making optimised policy decisions relative to the development of intermodal transport in Belgium, including environmental aspects and allowing the reduction of emissions in the transport sector.

Modelling and monitoring of pesticides fate and exposure in a regulatory context (P)

MOI19 Quantifying visual assessment of kinetics - Development of an objective criterion to support visual assessment of SFO fits of parent soil degradation J. Witt, Bayer AG / Environmental Safety; S. Boulke, Envisearch Ltd.; S. Ford, JSC International Limited; D. Patterson, Syngenta; B. Erzgraber, BASF SE; M.A. Thomas, Monsanto Company; I. Hardy, Batelle; R.L. Jones, K. Hammel, Bayer AG, Crop Science Division / Environmental Safety; J.A. Hingston, Chemicals Regulation Division

Kinetic evaluation of soil degradation studies for parent compounds is a key step to derive degradation endpoints. For modelling endpoints, single-first order (SFO) kinetics is preferred when acceptable, because it is implemented in exposure models. In presence of some bi-phasic tendency, acceptability of SFO is a recurrent source of discussions in the regulatory context. FOCUS kinetics guidance proposes Chi2err < 15% and visual assessment as decision criteria. However, the Chi2err margin leading as it does not account for systematic deviations, while visual assessment is inherently subjective. In the framework of a group led by UK CRD, to update FOCUS kinetics guidance, we aimed at finding criteria to quantify visual assessment. We collected 40+ example soil degradation studies that were assessed separately by 4 experts based on visual assessment, using scores between 0 (clearly SFO) and 10 (clearly bi-phasic). Individual scores showed high variability, preventing the deriving of the subjective visual assessment. Based on group discussions, we derived group consensus scores. Consensus scores showed little correlation with Chi2err (R^2 = 0.23). Among several proposed criteria, the SWARC (scaled and weighted area under the residue curve) criterion showed the best correlation to the consensus scores (R^2 = 0.77). SWARC was specifically developed for this task. The residue curve is split into blocks of the same sign (i.e., over- or underestimating the modelled data). The criterion is calculated as the root mean square weighted by the number of residues and summed up. The result is normalised by the study duration; a scaling factor accounts for high deviations from the last data
point. Thus, the criterion mimicks the visual assessment process, taking into account the presence and size of systematic deviations, and whether the model adequately predicts the last data point, as a measure for extrapolation capacity. We find that SFO fits with SWARC < 40 can be considered clearly acceptable; for one pesticide exceeded the threshold of 0.01 mg/kg. The soils also frequently contained multiple residues: 85% soils contained 3 or more pesticides and 51% soils 5 or more pesticides. Over half the soils (53%) contained at least 2 pesticides exceeding 0.01 mg/kg. The most frequent compounds were triazine herbicides (present in 89% soils), which were also in significant concentrations (47% soils with triazine sum exceeding 0.01 mg/kg). Based on the association with the occurrence of terbutylazine residues, it was concluded that this banned toxic simazine is still introduced significantly to the soils as an impurity of massively applied terbutylazine. Persistent atrazine residues are still a legacy of the past, even over 10 years after its last use. The second most frequent compounds were conazole fungicides (present in 74% soils; 53% soils with conazole sum exceeding 0.01 mg/kg). Although no health or environmental risk analysis has yet been carried out on the data, the results draw attention to potential impacts, because: (a) foreign limits based on risk calculations have often been exceeded; (b) many of these substances are suspected carcinogens or endocrine disruptors; (c) substances occur in mixtures whose (eco)toxicity may be additive or even synergistic. The research was carried out with the support of the GAČR (project 15-20065S).

MO122 Does the TOXSWA model simulate reliable concentrations in FOCUS surface water scenarios for a single segment water layer?

P.I. Adrianova, Alterra Wageningen University and Research Centre; W. Beltman, Alterra Wageningen UR

Recent development of approaches for quantitative use of surface water monitoring data in aquatic exposure assessments

W. Chen, Syngenta Crop Protection, LLC; P. Mosquin, J. Aldworth, RTI International

Current pesticide regulatory ecological exposure assessments conducted by the U.S. Environmental Protection Agency are almost exclusively based on standard scenario computer modeling. Monitoring data may exist from targeted (prospective or retrospective) programs and for general water quality research by industry, governments, and academic organizations. However, use of the monitoring measured data has been limited in the regulatory assessment process to refine/inform modeling. The limited use of water monitoring data is largely due to variability in the monitoring program sampling designs (frequencies, timing etc.) and insufficient information regarding the exposure conditions and the context sensitivity of the vulnerability of the monitoring location to a broader regional extent. In this paper, we summarize a set of recently developed approaches to infer and quantify realistic pesticide exposure potential based on monitoring data, including bias factor (BF), universal kriging (UK), and survey statistics. These approaches can be used in a systematic way to provide a useful reality check for comparison with exposure model output in regulatory assessments, thus increasing confidence in decision making. Examples of applying these approaches are provided to demonstrate their usefulness for watershed scale assessments.

MO123 Does the TOXSWA model simulate reliable concentrations in FOCUS surface water scenarios for a single segment water layer?

P.I. Adrianova, Alterra Wageningen University and Research Centre; W. Beltman, Alterra Wageningen UR

Bridging the hemispheres - Global use of field trials based on ecoregion similarities between New Zealand, Chile and Europe

B. Gottesbaeren, BASF SE / Crop Protection, Environmental Fate Modelling; H. Bayer, BASF SE; K. Platé, BASF SE Agrarzentrum Limburgerhof / Environmental Fate Modelling; B. Erzgraebner, BASF SE; F.P. Donaldson, BASF Corporation / APD/EPR; J. Goulet-Fontin, BASF SA; F. Kröger, Eurofins Agroscience Services GmbH

In European regulations degradation rates in soil (DegT50) from terrestrial field dissipation studies TFD studies considered for exposure modeling may originate from ”any” sites with soil and climatic conditions similar to Europe. An OECD Ecoregion similarity model (ENASIGPS) had been developed (OECD 2016) for gaining acceptance of field studies conducted in North America to Europe and vice versa, and there is no obvious scientifi
Multi-year evaluations in the FOCUS Surface Water assessment - results of beta testing

D. Weber, M. Brauer, Eurofins Regulatory AG / Environmental and Ecological Modelling; A. Boelehan, Bayer AG, Research & Development, Crop Science; G. Spickermann, ADAMA Deutschland GmbH; D. Schaefer, Bayer Crop Science / Environmental Safety


MO125 Spatial and temporal explicit catchment modelling in aquatic risk assessment using the modular framework CMF

S. Multsch, F. Krebs, S. Reichenberger, DR. KNOELL CONSULT GmbH; S. Heine, Bayer AG / Effect modelling; P. Kraft, L. Breuer, Justus Liebig University Giessen / Chair of Landscape, Water and Biogeochemical Cycles; T. Schad, Bayer AG / Environmental Modelling

The EFSA Guidance Document on Aquatic Risk Assessment indicates a key role for effect modelling in future aquatic risk characterisation in a tiered risk assessment framework. Such approaches require correspondingly adapted exposure tools and scenarios ranging from simple edge-of-field to spatiotemporally explicit landscape-scale catchment models. These approaches should be sufficiently flexible and transparent in order to design lower- and higher-tier scenarios of consistent protection. P vel. Nature 2015; 137: 1277-1279. Multsch et al. (2015) came up with a new model structure which makes adaption to different complexity levels possible. Flexible and modular approaches are needed to provide a spatially and temporally explicit aquatic exposure pattern to investigate effects on organisms according to Specific Protection Goals. A flexible and modular catchment model for water and pesticide transport has been developed which allows for stepwise adaption of model complexity to different assessment levels. The approach is based on the hydrological programming library CMF. Core functions of CMF are implemented in C++ and specific catchment setups are designed by Python scripting. The current approach focuses on the following abilities in order to investigate landscape-scale interactions: (a) a modular programming structure that enables replacement of process descriptions and (b) an incorporation of different spatio-temporal scales of terrestrial and aquatic processes into one model. This enables replacement of process descriptions and (b) an incorporation of different spatio-temporal scales of terrestrial and aquatic processes into one model. This approach allows for the explicit consideration of spatial differences in terrestrial processes and aquatic compartments. The model framework is currently being tested using a range of scenarios from simple edge-of-field to complex landscape-scale catchments.

MO126 Determination of runoff and drainage triggers for PEC surface water using automated simulation with FOCUS models

B. Kind, A. Guckland, T. Kleinmann, WXS Scientific GmbH

For the zonal registration in the EU predicted environmental concentrations in surface water need to be simulated based on the FOCUS models. Three different entry paths are considered: runoff (simulated in PRZM), drainage (MACRO) and spray drift (SWASH drift calculator). While the latter only depends on the amount sprayed, the distance to the water body and the spray equipment used, runoff and drainage amounts are also triggered by substance properties, e.g. degradation in soil and adsorption to soil. Often, a lot of runs need to be simulated for different crops or application timings to proof a safe use of plant protection products as defined in the Good Agricultural Practice (GAP). For this poster we evaluate the FOCUS scenarios compared to the substance properties DT50 and KOC. The idea is to find DT50 and KOC values which trigger runoff and drainage amounts and to distinguish worst-case FOCUS scenarios for different DT50 and KOC values. Dummy substances will be created which have different values for KOC and/or DT50 in soil. The remaining properties will be identical for each KOC/DT50 variation. Using automated FOCUS surface water simulations PECsw values were calculated for different scenarios at different application times within three risk assessment frameworks: runoff and spray drift as entry paths to focus solely on drainage and runoff. The results for different KOC/DT50 values of a single scenario were compared to identify a trigger value for runoff or drainage in this scenario. Furthermore, the results of different scenarios for a single substance will be compared to find the most sensitive scenario for these KOC/DT50 values. Finally, the amount of simulations necessary to show a safe use might be reduced to certain worst-case scenarios depending on the DT50 and KOC properties of the substance.
MO129
Recalibration and cross-validation of pesticide trapping efficiency equations for vegetative filter strips (VFS) using additional experimental data
S. Galimberti, DR. KNOELL CONSULT GmbH / Environmental Fate / Modelling / GIS; R. Sur, Bayer AG - Crop Science Division / Environmental Safety; C. Kley, Bayer AG Crop Science Division; S. Sittig, DR. KNOELL CONSULT GmbH / E-Fate Modelling; S. Miltusch, DR. KNOELL CONSULT GmbH

Vegetative filter strips (VFS) are widely used for mitigating pesticide inputs into surface waters via surface runoff and erosion. To simulate the trafficability of VFS in reducing surface runoff volumes, eroded sediment and pesticide loads the model VFSMART (Muñoz-Carpena and Parsons, 2014) is frequently used. While VFSMART simulates infiltration and sedimentation mechanistically, the reduction of pesticide load in surface runoff by the VFS (delha) is calculated with the empirical multiple regression equation of Sabbagh et al. (2009). This equation uses the following inputs: predicted reduction of total inflow (deltaQ) and eroded sediment load (deltaE), absolute surface runoff volume and eroded sediment load entering the VFS, linear adsorption coefficient Kd of the pesticide, and the clay content of the field soil (as a proxy for the clay content of the eroded sediment). The Sabbagh et al. (2009) equation, the coefficients of which were obtained by calibration against 47 data points, has not been widely accepted by regulatory authorities, on the grounds that its reliability has not been sufficiently established yet. Hence, evaluation against additional experimental data is necessary. Chen et al. (2016) proposed an alternative regression equation with a different structure based on 181 experimental data points. This equation uses fewer independent variables, but has more parameters than the Sabbagh equation. The objective of the present study was to improve the predictive capability of the Sabbagh et al. equation by breaking it down into subequations. For this, additional experimental data, for instance, concentration measured data were compiled from the available literature and thoroughly checked for their suitability. Moreover, existing errors in the calibration and validation data points of Sabbagh et al. (2009) were corrected. The consolidated experimental dataset (n = 244) was used to recalibrate the Sabbagh and Chen equations. Moreover, a sensitivity cross-validation analysis was performed to assess the predictive capability of both models. The Sabbagh equation fitted the whole dataset slightly better than the Chen equation (r² = 0.82 vs. r² = 0.79) and performed consistently better in the cross-validation exercise (with respect to the prediction performance indicators Q², predictive r², and RMSEP). Finally, a maximum-likelihood-based calibration and uncertainty analysis were performed for the Sabbagh equation using the DREAM algorithm.

MO130
Vanda - Visualize and Assess: a tool for the pesticide risk mitigation in surface water
F. Galimberti, G. Azzimonti, ICPS - International Centre for Pesticides and Health Risk; P. Rigoni, Public Health A. Moretto, University of Milan - The Directive 2009/126/CE of European Parliament and Council on Sustainable Use of Pesticides introduced a community action framework to protect the Environment of the EU and requested Member States to implement policies and actions in order to reduce the risk of pesticide use. In the region of Lombardy, in Italy, this Directive was adopted with DGR n. X/3233. The aim of the present work is to develop an interactive, easy to use tool to visualize the pesticidal load and water contamination, assess the potential pesticide risk and identify areas where to introduce mitigation measures to reduce the contamination, and consequently to reduce the risk in the surface water compartment. The datasets to start with are the monitored concentrations of pesticides in surface water, produced by the Regional EPA. These values are used in this context as Measured Environmental Concentration - MEC. The ratio MEC/PNEC is proposed in this work as a sort of risk assessment, even though the limitation and the complexity of usage of monitored data is well known. In addition, the ratio MEC/ESQ - Environmental Qualitative Standard (average annual concentration), is intended, to address the water quality with respect to the regulatory limit for pesticides in surface water (EEC 2000/60/EC). A MS Excel tool has been developed to map the monitored residues of pesticides, assess the potential pesticide risk (MEC/PNEC) and identify “hot spots”, that is areas where mitigation measures should be included. The tool is thought to be an anyone-can-use one, even with no particular knowledge of GIS or database management. Its peculiarity to be built inside MS Excel gives itself the possibility to share and to ease the dissemination of results. For more advanced mapping, the tool can interact with ESRI ArcGIS. The openness of Vanda makes it a tool suitable to work with other environmental compartments or other environmental theamatics.

MO131
Selecting application dates for UK higher tier drainflow modelling: comparing the FOCUS PAT and CRD PAT rules, and assessing the role of soil trafficability
J. Camall, G. Hughes, Cambridge Environmental Assessments; J.A. Hingston, J. Evans, Chemicals Regulation Division

Pesticide losses via drainflow are strongly dependent on the soil moisture status at the time of application and the rainfall pattern that follows application. For drainflow simulations, the choice of application date can therefore have a significant influence on predicted environmental concentrations. To standardise the selection of application dates, the FOCUS surface water models include a calculator tool, the Pesticide Application Timer (FOCUS PAT), which selects an application day on a calendar which has been defined by the user by applying a set of rules to the daily rainfall data used in the simulations. Alternative criteria for selecting application dates for drainflow simulations were proposed by Brown et al. (2004; Pest Manag Sci. 2004 Aug; 60(8); 765-76), and incorporated into a modelling tool developed recently by the HSE’s Chemicals Regulation Division (CRD) for performing UK higher tier drainflow assessments using the MACRO model. Under these rules (referred to herein as CRD PAT) a different set of criteria is applied to the daily rainfall data, with the algorithm selecting the first compliant date on or after a particular target day. Both the FOCUS PAT and CRD PAT algorithms select pesticide application dates based on daily rainfall volumes. In practice, however, application dates can be constrained significantly by the trafficability of the soil. Product GAPs are designed to cover a wide range of application periods to account for seasonal variation, e.g. in dry springs applications might take place in March while in wetter years, when it is more difficult to traffic wet soils, they may take place in April or even early May. When conducting multi-year modelling risk assessments this nuisance is often lost from the risk assessment, and applications are forced into months when farmers would not have been able to travel their lands and apply crop protection products. Using a soil moisture deficit based approach to pesticide application, the CRD and FOCUS PAT calendars consider different soil trafficability, and different algorithms were modified to account for this agronomic restriction. In this poster, the results from the four approaches – namely CRD PAT, FOCUS PAT, CRD Traffic PAT and FOCUS Traffic PAT – are contrasted and compared, with a view to drawing conclusions for the standard and refined UK higher tier drainflow risk assessment process.

MO132
Considering diffuse urban and agricultural sources of pesticides at the landscape and catchment scale
G. Hughes, J. Camall, Cambridge Environmental Assessments; F. Eriech, CEA for plant protection products (PPPs), there is a strong move towards landscape and catchment scale risk assessments as this allows for integrated risk assessments that consider multiple sources of pollutants, different exposure pathways as well as different receptors within a single framework. This landscape/catchment approach moves away from realistic worst case scenarios, designed to be protective of a wide range of usage situations, to a more realistic representation of usage environments through a risk assessment that is more relevant. Two important aims are to (i) develop a landscape/catchment approach to pesticide application, and (ii) build models to provide a new assessment approach. In this poster, we discuss the need for these two sources to be considered jointly within landscape and catchment scale risk assessments and the possible implications are discussed.

MO133
Calibration of passive samplers for the monitoring of chlordecone in French Caribbean rivers
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The uncertainty of the tropical weather in the French Caribbean makes the monitoring of chlordecone obsolete and new approaches should be explored to monitor the fate of this molecule. An aquatic system was chosen, where water is sampled under a continuous flow system, and the field calibration was done in triplicates in river Capesterre (Guadeloupe, French Caribbean). Rs in laboratory calibration were 0.30±0.02 L.day⁻¹ for the POCS1, 0.9±0.10 L.day⁻¹ for the POCS1n 0.1µm and 1.5±0.18 L.day⁻¹ for the POCS1n 0.01µm. Two distinct Rs have been calculated for the POCS1 and the POCS1n 0.1µm: one for the first five days of the experiment (R=0.10±0.01 L.day⁻¹ for POCS1; Rs=0.48±0.50 L.day⁻¹ for POCS1n 0.1µm, and one for the overall experiment (R=0.19±0.02 L.day⁻¹ for POCS1; R=0.43±0.01 L.day⁻¹). POCS1n 30µm followed the same pattern than in the laboratory calibration and reached equilibrium after 3 days, with a Rs significantly higher than
in the laboratory calibration (Rs=4.82 ±1.93 L g⁻¹). POCIS and POCISn samples can accumulate chlordecone efficiently despite its hydrophobic properties. POCIS 30μm seem to be a useful tool to monitor short flash floods, which happen regularly in this area.

MO134 Temporal patterns of pesticide residues in four major river basins in Korea C. Kim, K. Son, Y. Ihm, H. Lee, National Institute of Agricultural Sciences / Department of Agro-food Safety & Crop Protection
To evaluate residues of environmental concerned pesticides which mainly include pesticides used for rice cultivation, total ninety four sampling sites were selected through main streams and branch streams of four major river basins. And the water samples were collected at these sites from March to July-August and from July-September-October to November-December in 2012 and 2014. Besides, the water samples at sites of Keum, Mangyong and Dongjin rivers belong to the Keum river basin were regularly collected with a month interval, especially biweekly from May to August in 2013. Of the pesticides monitored, fenoxanil, hexaconazole, isoprothiolane, iprobenfos and thifluoximate as fungicides were mainly detected in rice season. While other fungicides including dimethconazole, propiconazole, fenamidone, nufamid and boscalid, were detected with low frequencies and their average residue levels in positive samples were also fairly low. Of the insecticides monitored, some organophosphorus, cadusafos, diazinon, fenitrothion, fenphos and prothiofos, two carbamates, carbofuran and fenobucarb, and endosulfan were detected with low frequencies and low residue levels. Surface water were collected at these sites, nine pesticides which include alachlor, butachlor, dimethametryn, dithiopyr, ethalfluralin, oxadiazon, simetryn and thiocthenbcarb were detected with frequencies of 1-48% and in their residue level of 0.01-1.9 µg L⁻¹ Detection frequencies and residue levels of insecticides and herbicides were the highest in waters sampled in May and June. Most pesticides detected were for the rice plants and their residue levels were very low to compare with standard values.

MO135 Occurrence of 14 representative pesticides in surface and ground waters of the State of São Paulo, the biggest sugarcane producer in Brazil R.D. Acayaba, SCHOOL OF TECHNOLOGY UNICAMP; C. Raimundo, UNICAMP / Institute of Chemistry; A. de Albuquerque, G. Umbuzeiro, School of Technology, UNICAMP / LAEG
São Paulo State is the biggest sugarcane producer in Brazil and the second at pesticide consumption. The aim of this project was to develop a method to determine the presence of 14 pesticides representative from sugarcane plantation, 7 herbicides (simazine, atrazine, ametryn, clomazone, diuron, hexazinone and tebuthiuron), 3 fungicides (azoxystrobin, carbendazim and tebuconazole), 3 insecticides (carbofuran, imidacloprid and malathion) and 1 transformation product (atrazine-2-hydroxy) in surface and ground waters using liquid chromatography tandem-mass spectrometry (LC/ESI/MS/MS) and solid phase extraction as sample preparation. Limits of detection (LOD) and quantification (LOQ) were ranged from 0.9 to 22 ng L⁻¹ and from 2.8 to 74 ng L⁻¹, respectively, and mean recovery was 66 %, which allowed obtaining a sensitive and accurate method for the determination at trace levels. In total, 196 samples located in the main sugarcane area from São Paulo were analyzed (175 surface waters and 21 groundwater) between October/2015 to October/2016. The most frequently detected pesticides in surface water were atrazine-2-hydroxy (100%), diuron (94%), carbendazim (93%), tebuconazole (92%), isoxaflutole (91%), imidacloprid (96%) and ametryn (81%). The pesticide that presented the highest concentration for this matrix was imidacloprid, reaching 2579 ng L⁻¹. The risk to aquatic life were evaluated dividing the maximum environmental concentration of each pesticide by the lowest water quality criteria found in the literature. The potential risk for aquatic life was observed for diuron, isoxaflutole, imidacloprid and malathion. For the groundwaters the most frequently detected pesticides were atrazine (90%), diuron (94%), carbendazim (91%), hexaconazole (99%), imidacloprid (96%) and ametryn (81%). The pesticide that presented the highest concentration for this matrix was tebuconazole, reaching 107 ng L⁻¹.

MO136 Exposure scenarios for aquatic risk assessment of pesticides in Brazil B. Jene, BASF SE / Environmental Fate; R.P. SCORZA JÚNIOR, Embrapa / EMBRAPA AGROPECUARIA OESTE; D. Máximo, R. Rebelo, IBAMA / DIQUA / CGASG; A.V. Waichman, Universidade Federal do Amazonas; N. Peranginangin, Syngenta Crop Protection, LLC / Product Safety; A. Tornisielo, Bayer AG / GENCS - E-Fate; L. Murakami, Bayer AG Crop Science Division; O. Perez-Ovillia, Bayer CropScience / Environmental Safety; H. Henry, Bayer / Environmental Safety; T. Haering, BASF SE.
A tri-partite technical working group consisting of regulators, academia and industry was formed to develop a framework for aquatic risk assessment of pesticides in Brazil. The framework should include a sophisticated science based approach resulting in a comprehensive guidance. The basis of the exposure assessment is the selection of the 90th percentile vulnerability which is seen to represent a reasonable worst case and is used as basis of the exposure assessment in other parts of the world. Water surface scenarios should be identified in six pre-defined climatic zones for the seven most important crops soybean, maize, sugar cane, wheat, cotton, citrus and coffee. Runoff and spray drift were found to be the main entrance pathways of pesticides into surface water bodies. Whereas spray drift mainly depends on technology and local climatic conditions during application, surface runoff is influenced by pedoclimatic conditions that could be assessed in a spatial vulnerability analysis. For this the USDA Runoff Curve Number approach (RCN) which is implemented in PRZM was used. A simple model based on the equations of the RCN approach was developed to calculate daily surface water runoff volumes for the agricultural area of Brazil for 34 climatic years. Calculations were carried out on highest available resolution of soil data resulting in more than 63,000 raster cells. Hydrological soil groups were determined by using a Brazilian specific classification scheme applied to the national soil map. Relative runoff vulnerability for pesticides was estimated with an integrated vulnerability index approach where indices for daily runoff such as average annual number of runoff events and average maximum runoff volume of each year were combined with an index for the expected substance concentration in the runoff water based on organic carbon content of the soil. The 90th percentile relative vulnerability was determined for the relevant crops for each climatic zone to select the relevant surface water scenarios. For the specific crop area, census data on municipality level were used first, but it was decided to switch to satellite images as fast as they become available. After discussions in the technical working group the Brazilian environmental authority IBAMA decided to use the US-EPA PWC model for the surface water exposure assessment. Representative flowing and static water bodies which need to be natural and permanent will be defined for each selected scenario.

MO137 Identification of Herbicide Source Areas and Spatial Variability of Dominating Transport Processes in a High Agricultural Intensity Catchment H. Rathjens, M.F. Winchell, Stone Environmental, Inc / Environmental Systems Management Inc; Sur, Bayer AG Crop Science Division; O. Baets, Bayer AG Crop Science Division / Sustainable Operations; F. Krebs, DR. KNOELL CONSULT GmbH; D. Lembrich, Bayer AG Crop Science Division
The occurrence of herbicides in surface waters of intensively cultivated catchments can originate from a variety of sources. These include transport via runoff and erosion during storm events, subsurface transport through lateral flow and through subsurface tile drainages, and from spray drift during applications. The Soil and Water Assessment Tool (SWAT) is widely used in the United States and the EU for catchment scale hydrologic and water quality modeling of non-point source chemicals in the environment. The SWAT model was applied to a 992 ha agricultural catchment in the Flanders region of Belgium to help in better understanding the sources of the herbicide detections observed in daily sampling over 3.5 years at two locations along the catchment’s primary stream. The SWAT model was calibrated to observed flow and chemical monitoring data, then used to characterize the relative contributions of herbicides via surface processes, subsurface processes, and spray drift. In addition, very vulnerable fields with significant contributions to surface water exposure were identified. A quantitative comparison between monitoring data and simulated exposure profiles was made to single out those high residue concentrations that could not be attributed to any of these traditionally considered exposure pathways, and could ultimately be only explained by point source contributions. The model results demonstrate that SWAT is capable of simulating streamflow in a small agricultural catchment, and is capable of simulating diffuse source pesticide concentrations. This allowed application of an approach to modelled monitored contamination distinguishing between diffuse source dominated high concentrations from those most likely affected by point sources. The SWAT model also proved useful in identifying the spatial variability in the dominant transport processes contributing pesticide residues to the stream. While surface runoff of soluble pesticide was the major non-point source contributor on most fields, lateral subsurface flow was found to be important as well, especially in the western portion of the catchment. Spray drift is likely the least significant contributor at the catchment scale. Overall, the analysis of monitoring data and modeling results shows that the potential for reducing herbicide concentrations in the study catchment can be addressed by mitigating both point source contributions from farmyards as well as diffuse sources.

MO138 Pesticides in water and surface sediments from Douro River estuary (Portugal) - assessment of environmentally relevant mixtures using acute toxicity bioassays C. Rocha, ICBRAS, U.Porto, CIMAR, CIMAR LA; C. Cruzeiro, CIMAR, CIMAR LA; Porto, CEF TUCT, U.Combra; S. Amaral, ICBRAS, Porto; E. Rocha, ICBRAS, U.Porto, CIMAR, CIMAR LA
The Douro River is an international water river that passes through extensive agricultural fields, of both Portugal and Spain, before reaching the estuary at Porto and Gaia cities. Therefore, the presence of pesticides is suspected. Accordingly, the evaluation of 56 pesticides of different categories (insecticides, herbicides, and fungicides) should be viewed as a priority for this habitat due to their negative impacts on the biota. For this purpose, water was collected over one year at six sampling sites in the Douro River estuary. Samples were extracted by solid-phase
MO139

Monitoring programme to investigate the presence of myclobutanil and its soil metabolite in Italian groundwater following use in pome fruit, stone fruit and vineyards

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Myclobutanil is a fungicide used mainly in pome and stone fruit areas, and in vineyards. For national registration in Italy, FOCUS groundwater modelling showed that the PECgw for its soil metabolite (X1129885; up to 6% of applied) reached up to 2 µg/L. This was accepted at EU level since the metabolite is not toxicologically relevant. However, according to national rules, this triggers a groundwater monitoring study for a metabolite when the PECgw is >0.75 µg/L. For completeness, myclobutanil was also monitored. To allow for a robust monitoring study, it was necessary to identify monitoring areas with the following characteristics: (i) be representative of an intensive use of myclobutanil, and (ii) reflect reasonable worst case scenarios for Italy. To facilitate this, a GIS-based indicator (PLI, Potential Leaching Indicator) was developed to integrate information about sales data, the spatial distribution of the target crops and the distribution of the FOCUS groundwater model scenarios throughout Italy. Use of the PLI allowed five suitable areas to be identified (Bolzano and Trento for apple trees, Verona for stone fruit and grapes, Forlì-Cesena for pear trees and stone fruit, and Matera for stone fruit). These areas are also representative of the FOCUS groundwater scenarios relevant in Italy (Châteaudun, Hamburg, Piacenza and Thiva). In each area, five monitoring wells were identified to cover a range of parameters (depth to groundwater, hydrology, pedology, and presence of the target crops). Subsequently, a three year monitoring campaign was conducted (autumn 2014-spring 2017). From a total of 150 samples analyzed, the concentrations of myclobutanil and its soil metabolite (X1129885) were below the LOQ (0.001-0.002 µg/L) in all cases and 94% of the samples analyzed in autunnil, the number of positive detections was 23 and of these, 20 samples showed concentrations well below 0.01 µg/L. Only in one sample did the measured concentration exceed the trigger value of 0.1 µg/L. However, a number of factors indicate that this is due to point source origin. Even better results were obtained for X1129885 where the number of positive detections was only 13, with concentrations less or very close to 0.01 µg/L. Based on these results it can be concluded that given the actual use conditions, the probability of myclobutanil and its soil metabolite exceeding the threshold value of 0.1 µg/L in groundwater in Italy is very low.

MO140

Identification of areas at risk of groundwater leaching in Italy for the fungimant 1,3-dichloropropene

R. Verro, University Milano - Bicocca - Lybra ambiente e territorio S.r.l. / Department of Earth and Environmental Sciences; R. Bradascio, Dow AgroSciences Italia srl / RD; C. Vaj, Dow AgroSciences Italia srl; A. Finizio, University Milano - Bicocca / Department of Earth and Environmental Sciences; C. Vaj, Dow AgroSciences EMEA B.V.; G. Hoogeweg, Fresenius GmbH; S. Reichenberger, DR. KNOELL CONSULT GmbH / Environmental Fate / Modelling / GIS

The 1,3-D is an active substance used worldwide in soil fumigant products for the control of cyst and free-living nematodes. It is used in a variety of crops including fruiting and vegetable crops. Soil sorption studies have shown that 1,3-D and its metabolites present characteristics of highly mobile molecules with a potential to leach into groundwater when applied in vulnerable areas such as sandy soil and in areas characterized by shallow groundwater. European member states have a wide range of groundwater monitoring activities for plant protection products and their metabolites, but analysis of 1,3-D and its metabolites is not currently part of the routine programme. In this study we illustrate a methodology allowing to identify areas most at risk where monitoring should be focused in priority, taking the example of Italy. The methodology considers three parameters: i) crops distribution, ii) soil properties and iii) 1,3-D use. The data, structured as layers of information are managed within a GIS, and are interacted to get the so called Uniform Geographic Units (UGU) which are areas of uniform in their characteristics and are representative of a specific set of values parameters. The data about the spatial distribution in Italy of crops where the 1,3-D is applied were first gathered from the last agricultural census, which provide information at a provincial scale. Successively, these data were refined at municipality scale. The identification of sandy soil areas in Italy was performed using pedological information extracted from different official sources. Subsequently, the information was used to identify the need of a distributed model to highlight the potential risks of this estuarine environment and of other comparable. The methodology considers three parameters: i) crops distribution, ii) soil properties and iii) 1,3-D use. The data, structured as layers of information are managed within a GIS, and are interacted to get the so called Uniform Geographic Units (UGU) which are areas of uniform in their characteristics and are representative of a specific set of values parameters. The data about the spatial distribution in Italy of crops where the 1,3-D is applied were first gathered from the last agricultural census, which provide information at a provincial scale. Successively, these data were refined at municipality scale. The identification of sandy soil areas in Italy was performed using pedological information extracted from different official sources. Subsequently, the information was used to identify the need of a distributed model to highlight the potential risks of this estuarine environment and of other comparable.
datasets were recommended. These recommended datasets, however, have been superseded by new datasets in the past few years. Specifically, daily weather and soils data have undergone significant updates, which are reflective of the considerable effort in Europe to update this spatial information. Not only does dataset choice, but also how datasets are being processed in a geographic information system, impact modelling results. Basic assumptions regarding aggregation of data, data slicing for determining climatic zones and data resolution influence our modelling results. In this poster, we will show the implications of data selection and data processing on a distributed modelling framework centered around GeoPEARL-4R. Specifically we will focus on differences between datasets, data set resolution, capturing variability and ones ability to model at the pan-European level within EFSA’s tier 3 guidelines.

MO144
Combining specific and public groundwater monitoring data as higher tier for pesticide regulatory risk assessment
A. Boivin, ANSES
Pesticides risk assessment for groundwater in France is performed according to Regulation 1107/2009. The European tolls are routinely used by considering the same models and same tiered approach. Groundwater monitoring data are identified as higher tier that may supersede modelling. Still, there is currently no agreed guidance available on the use of groundwater monitoring data for regulatory purposes. Work is ongoing on behalf the SETAC-EMAG GW group (www.setac.org/group/SEAGFest). The main issues when dealing with groundwater monitoring data is that they have only been little considered to cover were not seen as not being properly addressed. Monitoring programs have also been submitted and assessed at national level mainly to refine metabolite groundwater risk assessment. Combined information from targeted and public monitoring were keys to address the representativeness of monitoring programs. The French groundwater public network (wells) is vast and the database is available online (www.ades.eaudefrance.fr). This database (ADES) is owned by the BRGM (French Geological Survey). This database mainly active substances, more metabolites will be included in the future Proposals to combine targeted together with public groundwater monitoring data were made to enhance the representativeness of the GW monitoring conducted.

MO145
Minimal variation in input parameters highly influences PEARL and PELMO results: how can these results be trustable?
S. Ullucci, ICPS; L. Menaballi, International Centre for Pesticides and Health Risk Prevention
The calculation of Predicted Environmental Concentration of pesticides in groundwater (PECgw) is a crucial point in the registration and authorization process of plant protection products (PPPs) in Europe. Calculations are usually performed by FOCUS models, in particular PEARL and PELMO models. These models allow a realistic but conservative assessment of the potential leaching of pesticides in the groundwater compartment. Model results are influenced by substance specific parameters such as DT50, KOM and Freundlich coefficient (1/n). Great variations in PPE values are expected when high variabilities occurs in one or more of the parameters listed above. In this work, we demonstrate that PECgw outputs are significantly affected also by minimal variation of the same parameters. Considering that a minimal variation is intrinsic in all laboratory studies (es. 25% uncertainty in KOM determination was calculated applying the Horwitz equation), it is questionable whether a corresponding high variation in model is scientifically acceptable. In a previous project (York, 2017), dummy substances with different combinations of DT50, KOM and 1/n values were used in FOCUS PEARL, in order to quantify the influence of each single parameter on the final PECgw. It was verified that the sensitivity of PEARL model can be considered quite excessive. In this follow-up project, further calculations were performed using FOCUS PELMO to confirm the sensitivity of these two models, commonly used in a regulatory contest. Leachate concentrations were plotted as a function of KOM and as a function of degradation rate coefficient. PECgw obtained by the simulations of these two models were used to create a classification system for the input parameters KOM and DT50 according to models sensitivity. Conservative values for each parameter class, to be used in PECgw calculations, are proposed for all substances. This approach could minimize the effects of the intrinsic input variability providing a better scientific approach to the assessment of groundwater modelling in the regulatory context.

MO146
European regulatory network on pesticide groundwater monitoring
A. Grimsing, The Danish Environmental Protection Agency / Pesticides and Gentechnology; W. Koenig, UBA Umweltbundesamt; A. Boivin, ANSES; A. Poot, Cgpb; A. Schwen, AGES; M.E. Balmer, Plant Protection Chemistry; A. Massey, Chemicals Regulation Directorate; W. Tüting, German Federal Office of Consumer Protection and Food Safety
Groundwater monitoring data should be included in the assessment of the leaching risk of pesticides and their metabolites. Monitoring data is generated in most European countries, but their use for leaching risk assessment during pesticide authorization is hampered due to the following reasons: (i) The data is often not publicly available or available only in an aggregated form in a report, (ii) most data has not been available in the national language of the origin country, which makes it hard for other countries to use it for their own purposes and (iii) the interpretation of groundwater monitoring data requires detailed knowledge of the local geological, hydrological and climatic conditions, and also knowledge about crops and the use pattern for pesticides. To overcome these difficulties, a network among regulatory authorities in Europe involved in groundwater risk assessment of pesticides is proposed. The aim of the network is to exchange pesticide groundwater monitoring data and frequent updates about the focus of national monitoring, and to assist each other in the interpretation of the data. The network plans to have its first meeting in Copenhagen, Denmark, in September 2018 with an invitation to all interested European countries to participate. The poster will present the thoughts about the network and the status of the start-up, and aims at promoting the network to interested authorities.

MO147
Overview of measured wash-off factors from experiments suitable to derive a refined input for FOCUS modelling
G. Reinken, E. Hellpointner, Bayer AG, Research & Development, Crop Science / Environmental Safety; D. Sossalla, Bayer Crop Science / Environmental Safety
Recent regulatory interest in the wash-off process resulted in a proposal to the that the effects of wash-off should be considered as additional soil loading for FOCUS modelling of foliar applied pesticides (EFSA 2010, 2016). EFSA also proposed to increase the default wash-off factor for FOCUS modelling from 0.5 cm-1 to 1 cm-1 (EFSA 2012). On the other side, EFSA has stated that effects of wash-off should not be considered as additional worst case but rather as average effect (EFSA 2015, 2017). The wash-off factor of a compound is a product (formulation) specific modelling input parameter that can be experimentally determined. A generic experimental study design has been derived in a workshop organised by the European Crop Protection Association (ECPA). This study design was used for the experimental determination of wash-off factors for modelling purposes. Overall 25 individual wash-off factors have been determined experimentally, mainly under GLP. The data set comprises 12 formulations, 8 compounds and 6 crops. Experiments consider one heavy rainfall event of 15 mm over one hour, applied 24 hrs after foliar pesticide spray. The determined wash-off factors are normally distributed. The arithmetic mean of all single values is 0.38 cm-1 with a median of 0.40 cm-1. Just 7 of the 25 values are slightly above the existing default wash-off factor of 0.5 cm-1. The highest measured wash-off factor is 0.57 cm-1 and the lowest 0.14 cm-1. A wash-off factor of 1.00 cm-1 would be clearly outside the 3-sigma range of the experimental data set. This experimental evidence does not support the proposal to increase the existing default wash-off factor from 0.5 cm-1 to 1 cm-1. EFSA 2010: PPR Guidance 4093 - Predicting environmental concentrations in soil EFSA 2017: Guidance Document 4982 - Predicting environmental concentrations in soil

MO148
Leaching and plant uptake of trifluoroacetic acid (TFA) under cropped outdoor conditions
G. Reinken, M. Beckmann, Bayer AG, Research & Development, Crop Science / Environmental Safety; K. Hammel, Bayer AG, Crop Science Division / Environmental Safety; U. Köhrer, Bayer AG, Research & Development, Crop Science / Environmental Safety; M. Lamshoef, Bayer CropScience AG / R&D; S. Sittig, DR. KNOELL CONSULT GmbH / E-Fate Modelling
In recent regulatory discussions about the plant uptake process the question was raised if tracer-like substances (very low or zero sorption, very slow or no degradation) are fully available for uptake by plant roots under dynamic outdoor conditions. An alternative hypothesis is that such substances move so quickly in the soil column that they are taken up by plant roots to a lower extent than indicated by static laboratory plant uptake studies. To address this particular question an outdoor container study was conducted with 14C-labeled trifluoroacetic acid (TFA) and winter wheat plants for a time period of 228 days after application onto soil. Trifluoroacetic acid is a common breakdown product of several chemical product classes (e.g. refrigerants like HFCs and HCFCs used in air conditioning, pharmaceuticals, pesticides, pyrolysis of PTFE (used as coating in many products like Teflon® or GORE-TEX®)). Plant root uptake of TFA under static conditions has been determined in parallel with experiments using plants growing in nutrient solution (hydroponic study design). The translocation and uptake of trifluoroacetic acid observed in the outdoor container study was evaluated with the FOCUS model considering root uptake processes as routinely applied in standard FOCUS modelling. A transpiration stream concentration factor (TSCF) for TFA under cereal growing outdoor conditions could be derived. The results demonstrate that mobile, tracer-like substances are taken up extensively by plant roots even
under dynamic outdoor conditions; the hydroponic study design is suitable to
to determine conservative input parameters for regulatory modelling; uptake
experiments with cropped outdoor container may be suitable as higher-tier to
derive a refined TSCF. Further experiments will indicate to which extent this study
design is also suitable to derive refined TSCF for compounds with other sorption
and degradation properties.

MO149 Investigating the variance of edge-of-field deposits of spray drift
H. Holterman, Wageningen University & Research / Agrosystems Research; J. Van
de Zande, Wageningen University and Research / Agrosystems Research

Spray applications in arable crops often lead to off-target spray deposits downwind
from the treated field. Throughout several decades, many experiments have been
conducted by different researchers to quantify the downwind spray deposits.
Relations between downwind spray deposits and parameters like sprayer settings,
field conditions and environmental conditions were investigated. Still, there is a
large variance in the observed data that cannot be explained satisfactorily by the
experimental and environmental conditions. Sprayer boom movements and local
fluctuations in driving speed, wind speed and wind direction are the most likely
factors affecting variance in downwind spray deposits. In this study variations in
downwind deposits of spray drift caused by sprayer boom movements are
investigated both experimentally and based on simulations using the spray drift
model IDEFICS. Downwind deposits of spray drift were measured alongside a
treatment field at 2 m downwind. Consequently, the part of the spray that is applied
during the experiments. Horizontal and vertical movements of the sprayer
booms were recorded as well. Variance of spray deposits at 2 m downwind from the
field edge was about 50%. At 5 m downwind variance was about 50%.

A quasi-dynamic model was developed based on the IDEFICS spray drift model. In
the new model the effect of both horizontal and vertical boom movements on
downwind field spray deposits was studied. From the above mentioned experiments, the
most important frequencies and amplitudes of boom movements were derived.
Using these frequencies, the model simulations resulted in variances of spray drift
deposits similar to those established experimentally. Effects of fluctuating wind
directions are to be investigated in the near future.

MO150 Exposure assessment for edge-of-field watercourses next to tree nurseries
regarding spray drift deposits
H. Holterman, Wageningen University & Research / Agrosystems Research; J. Van
de Zande, Wageningen University and Research / Agrosystems Research

In the Netherlands about 90,000 live within 50 m of flower bulb or
fruit cultivation. It is unclear how many of these people are exposed to pesticides or
whether their health is at risk. Recently, a research project was launched to assess
the exposure of residents to pesticides next to flower bulbs fields. This research
projects involves both measurements and simulations of airborne spray drift.
Volatilization is investigated for several days after spray application. Outdoor and
indoor airborne concentration in tree nurseries involve spraying techniques that apply
the pesticide in upward or sideways direction. Particularly for high avenue trees the
downwind loss of pesticides due to spray drift can be relatively large. The upward
directed part of the spray that is blown towards the top of the trees may reach
heights above the trees, where wind can take the spray cloud and move it far
downwind. Usually, the branches and leaves at the lower part of the stems of high
avenue trees are cut away. Consequently, the part of the spray that is applied
sideways may pass underneath the tree canopies and reach downwind areas easily.
Measurements of downwind spray deposits for tree nurseries indicate deposition
levels comparable to those occurring for fruit orchards, which are well above those
occurring when spraying arable crops. The current paper deals with the
countrywide exposure assessment for pesticides applied to tree nurseries reaching
downwind field watercourses in the Netherlands. In high avenue trees, nurseries with the younger spindles and transplanted trees are considered as well. Although the total area of tree nurseries is limited, exposure risk
to edge-of-field watercourses is important enough to investigate. Spray drift
mitigation techniques are considered and evaluated as well.

MO151 Investigating the exposure of residents to pesticides due to airborne spray drift
H. Holterman, Wageningen University & Research / Agrosystems Research; J. Van
de Zande, J. Michielsen, H. Stallings, P. Van Velde, Wageningen University and
Research / Agrosystems Research

Flubendiamide belongs to diamide insecticide that has been used to control a wide
range of insects in fruit and vegetables. Assessment for agricultural applicator’s
exposure and risk is possible using exposure and risk assessment model developed by
EFSA called PRIMO (Pesticide Residue Intake Model) however its use is
limited to orchards. In addition in the case of polymers, from an analytical point of
view it may be difficult if not impossible to analyse the crops for residue content of
this type of co formulate. The objective of this work is to develop a methodology to
be applied under this conditions. As a case study we present this methodology for
latex polymers i.e. polymer based on methylene succinic acid with buta-1,3-diene,
styrene and methacrylic acid. [1] Prediction of agrochemical residue data on fruit
using an informatics system (PARDIS model), Calliera M, Balderacchi M, Capri E,
Trevisan M 2008

MO153 Dietary exposure to pesticide residues: the big picture
X. Baroza Bouza, L. Ferrer, EFSA - European Food Safety Authority /
Pesticides Unit
Science-based approaches and integrated risk assessments by using experimental
data, models for pesticide residues intake estimations, monitoring data considering
real exposure, etc. are working tools to contribute to the mission of the European
Food Safety Authority (EFSA) on protecting European consumers' health and the
environment in the field of postproduction residues. Maximum residue levels (MRLs)
are estimated by using a calculation model developed by EFSA called
PRIMO (Pesticide Residue Intake Model) based on the international agreed
methodology. This provides the key information to be interpreted by risk assessors and
for risk managers’ consideration. Essential input values in risk assessment include
toxicological data and residue values relative to many environmental scenarios and
considerations that are used to define and characterize the residues to which human
exposure can be estimated. The EFSA risk assessment procedure is based on the
upper levels of pesticide residues that are legally permissible in food of plant
and animal origin. Before an MRL is established, the EFSA assesses the residue
behavior of the pesticide and the dietary intake resulting from the residues
expected in food. The chronic and acute dietary consumer exposure to pesticide
residues is estimated by using a calculation model developed by EFSA called
PRIMO (Pesticide Residue Intake Model) based on the international agreed
methodology.
dermal exposure of flubendiamide was 3635.7 μg, while that of mixing/loading case was 815.3 μg. Hand exposure of flubendiamide (688.7 μg) in mixing/loading was higher than the case of application (680.8 μg). Exposure of body was highest (42.0%) in case of application. Penetration rate of insecticide between outer and inner dosimeter was about 0.5% (upper body) and 5.9% (lower body). Inhalation exposure during application was 20.2 μg while in case of mixing/loading no exposure was observed. Risk index (RI) was calculated to be 0.09 using 6 μg/kg/day of acceptable operator exposure level, suggesting that health risk of agricultural applicant during treatment of flubendiamide for cabbage field would be minimum. Keywords: Flubendiamide, exposure, risk assessment, whole body dosimetry, IOM, cabbage *Corresponding author: kj2404@snu.ac.kr; Tel, 82-02-880-4644

MOI55
Multi-focus Surface Water calculations: What do they mean for real regulatory cases? D. Schaefer, Bayer Crop Science / Environmental Safety; G. Reineken, Bayer AG, Research & Development, Crop Science / Environmental Safety; A. Bolekhan, Bayer AG, Research & Development, Crop Science; S. Heine, Bayer AG / Effect modelling; G. Goerlitz, Bayer CropScience AG / Environmental Safety The surface water exposure calculations for pesticides according to FOCUS are currently under revision by an EFSA working group. In particular the working group was mandated to extend the calculation period from a preselected single year to 20 years, with the objective to better compare the effect of variable weather conditions on aquatic exposure patterns. This is meant to provide a more robust and reliable basis for aquatic risk assessments in a regulatory context. The planned revision requires decisions on some technical aspects of the calculations (e.g. evaluation of the original FOCUSww weather data, filling of data gaps, completion of irrigation data sets, definition of multi-year application dates) and also new rules for the interpretation of the results. The main challenge in this context is the lack of extended weather data for the whole FOCUSww calculations. Surface water exposure is strongly driven by individual weather events triggering run-off or drainflow, and depends in a complex way on substance properties and use patterns. Since multi-year calculations are time-consuming, a systematic investigation of the consequences of the switch to 20 year calculations and associated changes of the preselection process is still missing. In this work we conducted such an investigation by running multi-focus FOCUS Surface Water calculations for several substances with a range of realistic properties and use patterns, and by analyzing the resulting exposure patterns. We discuss our experience with the calculations themselves, and present exposure characteristics of the different test substances. These allow some generic conclusions with regard to the consequences of currently discussed options for multi-year aquatic exposure calculations, and may support the technical and regulatory decisions that the EFSA working group has to take.

MOI156
Effectiveness of grass buffer strips in reducing Spinosad runoff S. Otto, Italian National Research Council, S. Gottardi, M. Passini, Agrea SRL; R. Bondi, AgroSciences Italia srl / RD; O. de Cirugeda Helle, Dow AgroSciences Outcome from FOCUS and a recent Document from Italian Ministry of Health allows vegetated buffer areas as a mitigation measure for runoff to surface water. The modelling framework includes default values relating to the pesticide removal efficiency of such buffers. Recent research suggests that these default removal efficiencies are not transferable to new runoff models. Surface water exposure is strongly driven by individual weather events triggering run-off or drainflow, and depends in a complex way on substance properties and use patterns. Since multi-year calculations are time-consuming, a systematic investigation of the consequences of the switch to 20 year calculations and associated changes of the preselection process is still missing. In this work we conducted such an investigation by running multi-focus FOCUS Surface Water calculations for several substances with a range of realistic properties and use patterns, and by analyzing the resulting exposure patterns. We discuss our experience with the calculations themselves, and present exposure characteristics of the different test substances. These allow some generic conclusions with regard to the consequences of currently discussed options for multi-year aquatic exposure calculations, and may support the technical and regulatory decisions that the EFSA working group has to take.

MOI157
EFSA’s innovative guidance on the establishment of the residue definition for dietary risk assessment R. Leuschner, EFSA - European Food Safety Authority / Pesticides, Regulated Products (REPRO); A. Friel, EFSA - European Food Safety Authority / Pesticides Regulated Products REPRO *The positions and opinions presented in this poster are those of the authors and are not intended to represent the views or scientific works of EFSA Commission Regulation (EU) No 283/2013 setting out the data requirements for pesticide active substances provides that the toxicological significance of compounds and their amount likely to be present shall be considered when judging which compounds are to be included in the residue definition for dietary risk assessment. The European Commission, EFSA prepared a guidance on the residue definition for dietary risk assessment which intends to complement the OECD guidance. The EFSA guidance is intended as a practical instrument helping risk assessors, on the basis of factual information (derived from toxicological and metabolism data), non-animal testing methods, by weight of evidence, to transparently: Conclude for which residues of a pesticide on food and feed commodities a hazard identification and characterisation is needed; Perform such a hazard identification and characterisation; Define the compounds that should be included in the residue definition for risk assessment. The innovative aspect of the EFSA guidance is a structured sequence of three modules, each of which addresses hazard characterisation and dietary exposure by selecting genotoxicity as the starting criteria for human health assessment. The modules are: Module 1: Exclusion of genotoxicity; Module 2: General toxicity assessment; Module 3: Decision making for residue definition for risk assessment. The guidance provides as appendices an analysis of ADI and ARfD distribution for pesticide active substances and three case studies illustrating the practical application of this modular approach to derive a residue definition for dietary risk assessment for isoproturon, spiroxamine and epoxiconazole. In September 2016, EFSA organised a technical meeting with stakeholders on its new guidance to exchange views. EFSA PPR Panel (EFSA Panel on Plant Protection Products and their Residues), 2016. Guidance on the establishment of the residue definition for dietary risk assessment. EFSA Journal 2016;14(12):4549, 129 pp. doi:10.2903/j.efsa.2016.4549. 2OECD (Organisation for Economic Co-operation and Development), 2009. Series on testing and assessment No. 63 and Series on pesticides No. 31 Guidance document on the definition of residue; ENV/JM/MONO(2009) 30; 28-Jul-2009. 3Info session on applications – pesticides - technical meeting with stakeholders on EFSA GD on residue definition for dietary risk assessment. http://www.efsa.europa.eu/en/events/event/160926

Alternative Approaches to Animal Testing for Ecotoxicity Assessments (P)

MOI58
Investigations on the bioconcentration of xenobiotics in the freshwater amphipod Hyalella azteca C. Schröder, Fraunhofer IME / Department Bioaccumulation and Animal Metabolism; J. Hollender, Eawag / Environmental Chemistry; Q. Fu, Eawag, Swiss Federal Institute of Aquatic Science and Technology / Department of Environmental Chemistry; V. Kosfeld, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology; D. Eisser, A. Schütte, Fraunhofer IME; J. Ebersbach, Fraunhofer IME, Institute for Molecular Biology and Applied Ecology; L. Bischof, Fraunhofer Institute for Molecular Biology and Applied Ecology IME. Bioconcentration factors (BCF) are needed for regulatory purposes to assess the bioaccumulative characteristics of a substance in the aquatic environment. Traditionally these BCFs are determined in fish flow-through tests according to TGD OECD 305. These fish bioaccumulation studies are time consuming, expensive, and demand many laboratory animals. Accordingly, alternative methods that replace, reduce and refine (3Rs) this test system are needed. Two promising alternative test approaches have been developed as alternative to in-vivo BCF testing: I) An invertebrate flow-through bioconcentration test system using the freshwater amphipod Hyalella azteca and II) in vitro depletion assays with rainbow trout hepatocytes or rainbow trout hepatocyte S9 fractions. Flow-through test with almost 20 compounds showed, that the H. azteca bioconcentration test could be an appropriate test to predict bioconcentration in the standard fish test. Bioconcentration studies with H. azteca would support laboratory animal welfare considerations using a non-vertebrate species, improve efficiency and reduce costs for BCF-testing. The results of the in vitro depletion assays have been successfully applied to improve in-silico predictions for BCF values by adding the highly variable impact of metabolic activities of the test organisms. In this study we present a total of five substances with different characteristics, four substances with log Kow values ranging from 2.5 to 4.5 and one ionic substance, were tested both test systems. To obtain a more detailed understanding of the metabolic activities in H. azteca, its metabolism of the five tested compounds were compared to the metabolites generated in vitro by rainbow trout hepatocytes. The results show that Hyalella BCF testing; in addition to in vitro assays and in silico predictions, may help to reduce, refine, and replace the classic BCF estimation with fish in accordance to regulatory needs.
MO159 Assessing Differences in Sensitivity to Aromatase Inhibitors Among Freshwater Fish Species

There is significant concern regarding potential impairment of fish reproduction associated with exposure to endocrine disrupting chemicals (EDCs). Aromatase is a steroidogenic enzyme involved in the conversion of androgens to estrogens. Inhibition of aromatase activity by exposure to chemicals can reduce levels of circulating estrogen leading to reduced synthesis of vitellogenin and production of fewer eggs by females. This mechanism has been extensively studied in the laboratory model species, fathead minnow (Pimephales promelas). However, differences in sensitivity to inhibition of aromatase among species of fish is largely unknown. This is particularly true for species that are not routinely studied in short-term reproduction assays, including many fishes of significant ecological and economic importance such as catfish (Ictaluridae), eel (Anguillidae), and perch (Percaidae). This study investigated in vitro inhibition of aromatase by the model inhibitor, fadrozole, across eighteen phylogenetically diverse species of freshwater fish. Concentrations of fadrozole that result in 50 % inhibition of in vitro aromatase activity were found to range from 0.40 to 6.08 nM among these species. The environmental relevance of this approach is confirmed by its adoption in the regulatory European framework for mercury measurement in fish. For other elements, the potential for cross-organism and cross-species comparisons is increased via percutaneous or intravenous injections or feeding. Subsequently, the animals were sacrificed, and internal organs were collected annually between 1990 and 2016. The results also support the use of sea turtle cell cultures as an ethical and reliable method for investigating toxicological effects of environmental contaminants.

MO161 Assessing differences in sea turtle organ sensitivity using cell-based toxicity assessments
K. Finlayson, Griffith University - Smart Water Research Centre / Australian Rivers Institute; F.D. Leusch, J. van der Merwe, Griffith University / Australian Rivers Institute

The long-lived nature of sea turtles and their lengthy residence in coastal foraging grounds can result in high exposure to contaminants from urban, industrial and agricultural sources. Organic or inorganic contaminants have been quantified in all species of sea turtles worldwide. However, very little is known about how these contaminants impact turtle health, valuable information for identifying populations at risk. In vitro exposure experiments using cell cultures established from turtle tissue provide an ethical, reproducible and cost-effective method to identify threats of environmentally relevant contaminants to sea turtles. In recent years, the majority of sea turtle cell lines have been established from skin samples. However, as ingestion is the main route of exposure for these animals, cell cultures established from organ tissues may be more pertinent. This study used a number of primary cell cultures established from the skin, ovary, heart, liver and small intestine from three individual green turtles to investigate the variation in sensitivity between organ types. Cytotoxicity of five organics and five inorganic compounds was investigated using a range of 113 cell lines, which were compared across a range of cell lines that will allow more robust and meaningful risk assessments to be conducted for sea turtles, assisting conservation and management strategies worldwide. Our results also support the use of sea turtle cell cultures as an ethical and reliable method for investigating toxicological effects of environmental contaminants.

MO162 Comparison of rat liver S9 to an animal-free alternative ewoS9R in the Ames fluctuation assay
J. Brendt, RWTH Aachen University; B. Thalmann, EWOMIS; K. Blum, University of Saskatchewan; K. Kauffmann, RWTH Aachen University / Department of Biochemical Engineering; S.E. Crawford, RWTH Aachen University / Institute for Environmental Research, Dept. of Environmental Analysis; A. Schiwy, EWOMIS; J. Büchs, RWTH Aachen University / Department of Biochemical Engineering; H. Hollert, RWTH Aachen University / Institute for Environmental Research

The Ames test is the most important in vitro test for mutagenicity performed in many variants. The original agar-plate assay was modified to reduce the amount of assay components by using a total of 13 cell chord and differences between cell cultures were investigated based on their order of sensitivity to all compounds tested. The results of this study are being used in the construction of a cross-species quantitative structure-activity relationship (qsar) of Bioaccessibility-activity data as a relative-sensitivity adjustment for prediction of impacts at the individual and population level. This information could guide more objective ecological risk assessments of native species to EDCs that inhibit aromatase. The content of this presentation neither constitute nor necessarily reflect US EPA policy.

MO160 Fish scales as a tool for temporal biomonitoring of trace element concentrations
D.A. Vignati, CNRS / LIEC UMR7360; G. Masson, Université de Lorraine and CNRS / LIEC UMR7360

Direct measurement of contaminant concentrations in biological tissues is attractive for regulatory purposes because it accounts, in principle, for the environmental factors controlling their bioaccessibility and bioavailability. In the case of trace elements, the validity of this approach is confirmed by its adoption in the regulatory European framework for mercury measurement in fish. For other elements, the current framework privileges measurements in the dissolved (filtered) aqueous matrix. However, this approach provides only a limited temporal resolution of possible trends in elements’ concentrations and, where relevant, neglects uptake via dietary pathways. Furthermore, when fish is the selected matrix for monitoring, one or more individuals have to be sacrificed to collect the material necessary for analysis; a strategy that, apart from the associated ethical problems, may become problematic in situations where the number of resident fishes is limited. In this context, we evaluate the use of fish scales as a non-lethal, rapid and efficient alternative for monitoring trends in trace element levels in a reservoir receiving cooling waters from a nuclear power generation plant. The variations in the concentrations of Cu, Zn and lanthanides were followed in fish scales from archived fish material (Abramis brama) collected annually between 1990 and 2016. Scales were dried, calcined and mineralised using concentrated nitric acid. After digestion, Cu and Zn were assayed by atomic absorption spectroscopy and lanthanides by ICP-MS. For Cu and Zn, triplicate measurements generally agreed to 10% and accuracy, with respect to the standard reference material TORT2 (lobster hepatopancreas), was better than 90%. In the case of lanthanides, analysis were made on one sample per year and only La, Ce, Pr, Nd, Pm, Sm, Eu and Gd could be systematically quantified. Overall, fish scales proved to be a suitable matrix for the biomonitoring of trace element concentrations, including elements of emerging concerns such as lanthanides. Compared with fish muscle (or whole fish), fish scales are easier to preserve (no need for dissection, freezing and lyophilization) and, in some situations, may even allow repeated surveillance of the same individuals. This methodological validation study paves the way to further research to establish relationships between accumulation in scales, internal organs and biological responses.

MO163 QSAR: a predictive approach for electronic cigarettes toxicological assessment
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Department of Theoretical and Applied Sciences (DiStA); E. Caruso, University of Insubria / DISTA; S. Zucchi, S. Sterpone, E. Ferri, TRUSTICERT SRL
Electronic cigarettes (e-cigarettes) are devices that typically deliver nicotine, flavorings, and other additives to users via an inhaled aerosol. They are designed to closely mimic the experience of smoking conventional cigarettes. Nowadays, e-cigarettes are the most commonly used tobacco-related product among youth, surpassing conventional cigarettes in 2014. However, insufficient data are available to fully predict the exposure with the aerosol to which consumers are exposed; at the same time studies evaluating whether e-cigarettes are less harmful than cigarettes are inconclusive. Minimal valid chemistry data are available on e-cigarette emissions and no standardized methods and threshold values exist for e-cigarette analysis. To fill the chemical and toxicological data gap, comprehensive assessment of e-cigarette emission and toxicological studies are certainly needed. The aim of the present study is to provide, by means of Quantitative Structure-Activity Relationship (QSAR) approaches, a first toxicological screening of several e-liquids ingredients. Different Ordinary Least Squares (OLS) regression-based QSAR models were developed to define the potential acute toxicological profile of 265 molecules contained into e-liquids. The chemicals were identified as DCF, LD50 mouse and oral L50 in mouse and rat respectively. Theoretical descriptors were calculated by PaDEL-Descriptor software, and the best modelling variables were selected in the software QARINS. Models were validated for robustness, stability and absence of chance correlation using leave-one-out, leave-more-out and the scrambling of the responses. External validation was performed on multiple external prediction sets. This work was supported by the Research on Regulatory Science of SETAC Europe 28th Annual Meeting Abstract Book

enormous diseases. Epidemiological evidences show that the disease is associated with exposure to hazardous chemicals. However, causal relationship has not been clearly understood. In the present study, we aimed to elucidate the link between occurrence of metabolic or neuro disease and exposure of environmental chemicals. We first screened potential of environmental chemicals on the disease model organisms, C. elegans and Zebrafish. To maximize the advantage of these model organisms and reduce the cost of screening, using C. elegans mutant: oga-1(ok1207), ogt(ok1474), n1(ok259), transgenic zebrafish, Tg(T2Kins:nlsB-mCherry) and Tg(elav13:EGFP)kn3.

The highly conserved O-GlcNAc transferase and O-GlcNAcase. OGA is a gene that is related to type 2 diabetes and null mutations cause alterations in C. elegans carbohydrate and lipid metabolism. Neurogliin NLG-1 control synaptic function, which is conserved from nematodes to mammals and is needed to attend to high-intensity activity disorder (ADHD).

Assessing the bioaccumulation potential of several pharmaceuticals using fish

The use of plant protection products has exponentially increased in the agricultural sector over the past decades. Copper sulfate and glyphosate are two commonly used pesticides, the former as fungicide and the latter as herbicide. Farm animals may be exposed to this type of products through different ways: i) the drift of pesticides during their application may lead to inhalation or dermoc skin contact or ii) through the incorporation of pesticides. CMT/MIT, PGIH, were screened using C. elegans reproduction assay and zebrafish transgenic assay. The primary results showed CMT/MIT and BPA reduced fluorescence intensity of insulin gene in zebrafish, suggesting possible involvement of these chemicals on metabolic pathways. In brief, our results suggest multi-model approach could complement the limitations of a stand-alone model organism and thus more accurately identify chemical hazard on human health. Therefore, the outcome of the current study could be utilized for efficient chemical screening and better risk assessment of the chemicals. Acknowledgement: This work was supported by the Mid-career Researcher Program (2017R1A2B3002242) through the National Research Foundation of Korea (NRF) funded by the Ministry of Science, ICT and Future Planning.

In vitro effects of two pesticides on the motility and viability of bovine spermatozoa

The use of plant protection products has exponentially increased in the agricultural sector over the past decades. Copper sulfate and glyphosate are two commonly used pesticides, the former as fungicide and the latter as herbicide. Farm animals may be exposed to this type of products through different ways: i) the drift of pesticides during their application may lead to inhalation or dermoc skin contact or ii) through the incorporation of pesticides. CMT/MIT, PGIH, were screened using C. elegans reproduction assay and zebrafish transgenic assay. The primary results showed CMT/MIT and BPA reduced fluorescence intensity of insulin gene in zebrafish, suggesting possible involvement of these chemicals on metabolic pathways. In brief, our results suggest multi-model approach could complement the limitations of a stand-alone model organism and thus more accurately identify chemical hazard on human health. Therefore, the outcome of the current study could be utilized for efficient chemical screening and better risk assessment of the chemicals. Acknowledgement: This work was supported by the Mid-career Researcher Program (2017R1A2B3002242) through the National Research Foundation of Korea (NRF) funded by the Ministry of Science, ICT and Future Planning.

MO168

In vitro effects of two pesticides on the motility and viability of bovine spermatozoa

I. Bulhosa, University of Aveiro / Biology department; M. Lopes, ICBAS-University of Porto / Department Veterinary Clinics; I. Lopes, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro

The use of plant protection products has exponentially increased in the agricultural sector over the past decades. Copper sulfate and glyphosate are two commonly used pesticides, the former as fungicide and the latter as herbicide. Farm animals may be exposed to this type of products through different ways: i) the drift of pesticides during their application may lead to inhalation or dermoc skin contact or ii) through the incorporation of pesticides. CMT/MIT, PGIH, were screened using C. elegans reproduction assay and zebrafish transgenic assay. The primary results showed CMT/MIT and BPA reduced fluorescence intensity of insulin gene in zebrafish, suggesting possible involvement of these chemicals on metabolic pathways. In brief, our results suggest multi-model approach could complement the limitations of a stand-alone model organism and thus more accurately identify chemical hazard on human health. Therefore, the outcome of the current study could be utilized for efficient chemical screening and better risk assessment of the chemicals. Acknowledgement: This work was supported by the Mid-career Researcher Program (2017R1A2B3002242) through the National Research Foundation of Korea (NRF) funded by the Ministry of Science, ICT and Future Planning.

MO169

Assessing the bioaccumulation potential of several pharmaceuticals using fish S9 and hepatocyte assays

L.A. Constanting, Pfizer, Inc. / PDM; M. Embry, ILISI, R. Sharma, Pfizer / PDM Aquatic Toxicology for Environmental Assessment, Helmholtz Centre for Environmental Research, Helmholtz Centre for Environmental Research - UFZ / Department of Ecological Chemistry

MO170

Chemoavailability of Organic Electrophiles - A Nonanomial Approach to Identify Candidates for Reactive Toxicity

A. Böhme, UFZ - Helmholtz Centre for Environmental Research / Ecological Chemistry; G. Schuithämm, Helmholtz centre for environmental research - UFZ / Department of Ecological Chemistry

Organic electrophiles are important components within the exposomes of humans, flora and fauna. Their toxicity toward aquatic organisms is driven by two molecular initiating events (MIE): the hydrophobicity-triggered disturbance of cellular membranes and the chemical reaction with nucleotides, proteins, peptides or the DNA. The toxicity enhancement T, which indicates the ratio of narcosis baseline (hydrophobic MIE) vs. experimental in vivo or in vitro bioassay toxicity, has been used as a measure for the reactive MIE for many years. However, very early studies already showed that T does not solely depend on reactivity, but also decreases with increasing hydrophobicity. This indicates that the relevant hydrophobic targets are located in aqueous compartments and that the hydrophobic and the reactive MIEs do not contribute independently to overall toxicity. In this communication, we employ our concept of chemoavailability to a set of 58 Michael acceptors, in order to analyze the impacts of reactivity and hydrophobicity on the overall toxicity as well as on T. To this end, reactivity was quantified by the second order rate constant for the reaction of the Michael acceptors with glutathione (GSH). The hydrophobicity through the octanol/water partition coefficient and toxicity through the 48-h-effect concentration yielding 50 % growth inhibition of Tetrahymena pyriformis. The results demonstrate that the decreasing T with increasing Km is caused by a rate-determining transfer-step of the electrophile from lipophilic compartments into the aqueous cytosol. Finally, chemoavailability, as a trade-off between log Kow and log Km, is shown as a promising non-animal tool to analyze whether an aquatic toxicity is predominantly driven by the hydrophobic or the reactive MIE, or by both MIEs working in parallel. The authors thank the EU-funded project OSIRIS (GOCE-CT-2007-037017) and the BMBF-funded project ProHapTox (FKZ 031A422A and 031A422B) for financial support. [1] Mulliner D, Schüürmann G 2013. Mol. Int. 32: 98-107. [2] Böhme A, Lapa A, Schüürmann G 2016. Chem. Res. Toxicol. 29: 952-962. [3] Läpple A, Thaens D, Paschke A, Schüürmann G 2009. Chem. Res. Tox. 22: 742-750.

MO171

Local Electrophilicity Describes Experimental Glutathione Reactivity and Aquatic Toxicity toward Tetrahymena pyriformis

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Electrophilic compounds such as afl-unsaturated carbonyls are valuable reactants in organic synthesis and commonly used as industrial intermediates and products. At the same time, their electrophilic reactivity may become critical in case of uptake into organisms because of the ubiquitous presence of nucleophilic sites in proteins and DNA, resulting in reactivity driven excess toxicity. Therefore, exposure to electrophiles is of high toxicological concern. Thus, identification of toxicologically relevant compounds is desired. A step forward would be to predict - rather than measure – the electrophilic reactivity of compounds directly from molecular structure. This would enable screening with regard to their intrinsic toxicity potential. In this work, local electrophilicity parameters were developed based on the quantum chemical structure descriptors. The local electrophilicity of reactive toxicity was investigated. For this purpose, their correlation with logarithmic reaction rate constants toward Gluthathione (GSH) was analyzed. GSH is a small tripeptide which acts as a protector against electrophiles in the cytosol. The dataset of electrophilic chemicals contained 97 afl-unsaturated esters, ketones and aldehydes. In the context of aquatic toxicity toward Tetrahymena pyriformis, reactive toxicity is assumed to be the primary mode of action of the aforementioned compound classes. Therefore, the descriptive power of calculated and experimental GSH reaction rate constants was compared. Both models perform equally well and yield root mean squared errors of about 0.4 log units in modeling the toxicity enhancement as deviation from narcosis level based on 48h-inhibition-growth concentrations.

MO172

Using mechanisms of toxic action to classify and predict ester ecotoxicity

P. Bicheng, P. Bauer, KREATIS; P.C. Thomas, CEHTRA SAS / Ecotoxicology and Risk Assessment

Even though esters are often used and released into the environment, little is known about the mechanistic basis of their toxic action. Therefore, the poorly soluble esters are considered to exert a specific narcosis, while some other esters can exert toxicity related to their potential reactivity. Therefore the critical step, before predicting the toxicity of an ester, is to determine its mechanism of toxic action (MechoA). For this purpose the classification of Bauer et al., (2018) is used in combination with an accurate modelling approach which is derived from empirical data specific to the MechoA. The acute toxicity of esters to aquatic flora and fauna may be assessed against a hydrophobicity descriptor (i.e. log Kow or water solubility), and compared with similar regressions for non-polar narcotics. The similarity between these regressions confirms non-reactive esters are simple
narcotic compounds for algae but not for fish or daphnids. For the animal species, the regressions for esters are not the same as for narcotic compounds. The most likely explanation for this difference is the balance between hydrolysis rate and the toxic action of the parent and the degradation products. That is why they are considered as pronarcotics. A toxicity to algae in line with a narcotic mode of action suggests that the enzyme responsible for hydrolysis found in fish and daphnids is absent or therefore hydrolytic activity of esters is negligible. The di-esters appear as toxic than mono-esters for fish and daphnids because they can produce two times more metabolite than mono-esters. The more reactive esters are usually unreacted, like alkyl/vinyl-esters and alpha,beta-unsaturated esters, whose double bond can be activated by the carbonyl group. For these compounds the substitution around the double bond plays a decisive role in effective reactivity. Thus, methacrylates which have an alkyl substituent in position alpha of the carbonyl are not more toxic than aliphatic esters for any of the three aquatic species. On the other hand, Acrylates clearly express excess toxicity and have to be considered as acting through yet another mechanism of action relating to a model dedicated to soft electrophiles. Rather than model the toxicity only according to structural analogy, a modelling approach is used to develop QSARs for esters based on three pillars: structure, mechanism and species metabolism.

MO173 Nanosecond pulsed electric field incorporation technique to predict molecular mechanisms of teratogenicity and developmental toxicity on fish embryos
K. Arizono, Prefectural University of Kumamoto / Faculty of Env. Symbiotic Science; A. Yamaguchi, National Institute of Technology, Ariake College; M. Ishibashi, Ehime University / Faculty of Agriculture; S. Kono, National Institute of Technology, Ariake College; N. Tominaga, Ariake National College of Technology / Department of Chemical and Biological Engineering
We developed and applied the nanosecond pulsed electric field (nsPEF) treatment condition and assessed the teratogenicity and embryonic developmental toxicity by chemicals using fish embryos. Furthermore, we analyzed gene expression profiles in fish embryos using DNA microarray and performed pathway and network analyses to understand the molecular mechanisms of chemicals in teratogenicity and embryonic developmental toxicity. Our findings suggested that the PEF technique is a powerful tool for assessing teratogenicity and embryonic developmental toxicity of chemicals and predict their molecular mechanisms in fish embryos.

MO174 Moving 3D in vitro intestinal models forward: transcriptional characterization of the RtgutGC cell line
L.M. Langan, Plymouth University / Biological and Marine Sciences; S. Owen, AstraZeneca / Safety Health Environment; A.N. Jha, Plymouth University / Biological Sciences
Intestinal derived cell lines are useful in vitro models which allow for focused investigation of xenobiotic responses in the intestine. The establishment of the first immortalized intestinal cell derived from the rainbow trout (RtgutGC) offer an opportunity to explore intestinal uptake without the need for the use of numerous animals. Recent work using numerous compounds has acknowledged its potential as a replacement tool for animal based laboratory studies, there is still a lot to be explored before its widespread incorporation as a toxicology tool. Cell lines are known to acquire additional mutations or modifications while in culture, and it is important to understand to what extent this cell line retains the genetic landscape of primary intestinal tissue. In this study, RNA-Seq sequencing of the RtgutGC cell line was used to establish gene expression in this potential animal replacement model. Over 84% of the sequences were mapped to the genome. Following filtering for transcript abundance using TPM (transcripts per million), 24,890 contigs were identified and blasted against the NR database. InterProScan was run in parallel to blast annotation and later merged with annotation to confirm. Over 43 genes were shown to be differentially expressed in the cell line compared to the native tissue while 229 were shown to be down regulated. KEGG pathway analysis revealed the presence of significant metabolism pathways still active in the model. This study provides the first in-depth sequencing data of any rainbow trout cell line and identifies many commonalities between the 3D model and native tissue. Characterization of the RtgutGC transcriptome and genes and enzymes expressed in this model will greatly help in building realistic in silico models of exposure when integrated with other available chemical data.

MO175 Impact of test concentration on the in vitro intrinsic clearance using trout liver S9 fractions to predict the bioaccumulation potential of fragrance chemicals
Bioaccumulation in aquatic species is a critical endpoint in the regulatory assessment of chemicals. The bioconcentration factor (BCF) is usually determined in fish (OECD TGL 305). In vitro systems measuring biotransformation rates of chemicals to refine BCF model estimates have been established as alternative methods to refine predictive models which are based on hydrophobicity (i.e. log Kow). Fragrance chemicals frequently contain different isomers complicating its analysis especially at low concentrations. Thus, they have been commonly tested at 1 µM. Results reported recently indicate that first order depletion rate constants (kdep) measured at test concentrations of 1 µM could underestimate the in vitro intrinsic clearance resulting in overestimation of the BCFs. However, these observations were mainly reported for substances from one chemical class (polyaromatic hydrocarbons, PAHs). For pyrene, chrysene and benz(a)pyrene, kdep determined at lower concentrations were 4- to 12-fold higher than kdep measured at 1 µM. However, the effect of test concentration of industrial chemicals is lacking. The goal of this study was to compare kdep values using different concentrations (e.g. 2.2, 1 and 5 µM) for four fragrance chemicals. These chemicals represent a diverse class of high log Kow (4-3.6-5) industrial chemicals. Rainbow trout liver S9 fractions from different sources were used and their enzymatic activity characterized using commonly used fluorescence assays (EROD, p-nitrophenol glucuronidation and CDNB-glutathione conjugation) and substrate depletion assays with testosterone, 7-hydroxycoumarin, pyrene and Cyclohexyl salicylate as reference chemicals. Decrease of the parent chemicals was analysed by GC-MS or LC-MS and kdep values determined. For the lowest concentration (0.2 µM) ca. 2-fold higher kdep values were observed for Polysantol, Ambroxol, Cyclohexyl salicylate and Karanal compared to kdep values determined with 1 µM. Measured kdep values were 2-fold lower with 5 µM except for a 4-fold lower rate for Polysantol compared to 1 µM test concentration. The biotransformation rates of the fragrance chemicals tested were not significantly different from those of reference chemicals (0.2-5 µM) compared to PAHs indicating that their Kow may be substantially higher. Thus, for fragrance chemicals which are moderately to rapidly biotransformed, the use of 1 µM as start concentration seems to be a suitable approach to estimate the bioaccumulation potential.

MO176 Biological effects of 3 metals on "D" larvae of Japanese oyster Crassostrea gigas
A. Sobrino-Figueroa, Universidad Autónoma Metropolitana Iztapalapa / Hydrobiology; C. Cárdenas-Martínez, Universidad Autónoma de Baja California Su
The Japanese oyster is an introduced species from Asia, which is cultivated in the coastal systems of the Mexican Pacific. Due to the fact that in the last 10 years the populations have had problems in their survival, in this work the evaluation of 3 biomarkers was performed in "D" larvae of this species, exposed to the metals Cd, Cr, Pb and its mixture, because these xenobiotics, are in high concentrations in the sites where the oysters are grown. Bioassays (72 hrs) were conducted where the "D" larvae were exposed to the different concentrations of metal and their proportions in proportion: 1:1. With the obtained data, the LC50 was calculated and the evaluation of 3 biomarkers was measured in the surviving organisms: the degree of lipoperoxidation (TBars: Buege & Aust. 1978), the activity of the AcChE enzyme (Eltman et al., 1961) and genetic damage ( Comet Test: Singh et al., 1988). The toxicity of metals according to the calculated LC50 values was: (from most to least toxic): Pb > Cd > Cr. The comet test was used to assess the genotoxicity of the reference chemicals. Decrease of the parent chemicals was analysed by GC. For the metal mixture (0.2 µM) the comet test revealed a significant decrease in the activity of the AcChE enzyme and genetic damage between the exposed organisms and the control group. The metal with the highest oxidative effect was Chromium (32 ± 8.97 µM TBars mg⁻¹). And the metal mixture: Cd + Cr + Pb (45 ± 11.89 µM TBars mg⁻¹). In the evaluation of genotoxicity it was observed that Chromium had the highest effect (91% cells with damage) and Lead the lowest (43%). Cadmium and the mixture of metals caused inhibition in the activity of AcChE (56% ± 38% respectively). The results of this study show that the Cd, Cr and Pb metals in sublethal concentrations have deleterious effects on the "D" larvae of Crassostrea gigas.

MO177 Toxicity effects caused by exposure to Dichlorvos in organisms of different trophic levels
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Dichlorvos (DDVP) is an organophosphate insecticide considered by the EPA as highly toxic. Because there are few studies of the effects of DDVP in aquatic organisms The objective of this study was to evaluate the toxicity of Dichlorvos in organisms of different trophic levels Cladocerans: Daphnia magna, Daphnia exilis, Daphnia pulex and Simocephalus musellus, The ostracod Cypris sp. and fishes: Fundulus heteroclitus (Chromis chromis jordani) and juvenile zebrafish (Danio rerio). In addition their sublethal effects were evaluated by means of assessment of four biomarkers (growth rate, O.N index, liperoxidation and inhibition of acetylcholinesterase enzyme). Acute bioassays were performed, the organisms were exposed to 6 pesticide concentrations to determine the LC50. Subsequently tests with duration of 15 days were made where the organisms were exposed to a sublethal concentration (LC10, for assessment of 4 biomarkers (growth rate, O.N index, lipid peroxidation and inhibition of acetylcholinesterase enzyme). The LC50 values obtained in the bioassays varied from 5.300 to 0.021 mg L⁻¹. In the tests it was evident that the cladoceran Daphnia exilis was more sensitive to DDVP compared
provide evidence for the ease and potential application of this technique for future environmental monitoring programs. Our ongoing method refinement and exploration of this technique in sentinel fish with diffuser livers across different size ranges will strengthen our goal of proposing ultrasound as a viable non-lethal alternative to measure HSI on a global scale.

**MO180**

**Weight of evidence for fish acute toxicity: a Bayesian network modelling approach**

J. Mogsgaard, Norwegian Institute for Water Research (NIVA) / Section for Catchment Processes; A. Lillicrap, NIVA / Ecotoxicology and Risk Assessment; W.G. Landis, Western Washington University / Institute of Environmental Toxicology; S.E. Belanger, The Procter & Gamble Company / Environmental Stewardship and Sustainability Organization.

Reduction of animal testing wherever possible is requested by EU Directive 2010/63/EU. Fish Embryo Toxicity (FET) testing can be an alternative to using juvenile fish in acute toxicity testing. However, FET data are currently not accepted as a replacement to juvenile fish acute toxicity data for regulatory purposes such as REACH, without sufficient weight of evidence (WoE). The development of a WoE approach for FET data has been recommended by the European Chemicals Agency to significantly reduce the number of animals required for hazard assessments of chemicals. We propose a Bayesian network (BN) modelling approach for quantifying the weight of evidence. BN is a probabilistic modelling methodology which is an increasingly used in ecological risk assessment as well as in many other fields.

**MO178**

**Characterising estrogenic activity of arctic char tissue extracts in two fish in vitro models**


Contaminants from anthropogenic activities find their way to the Arctic through long-range atmospheric transport, ocean currents and via transport by living organisms (migrating fish or seabirds). Although the levels of POPs in arctic fish are generally low, local hot-spots of contamination have been demonstrated in freshwater systems affected by seabird guano, such as Lake Ellasjøen at Bear Island (Norway). High concentrations of organic halogenated compounds have been measured in resident populations of Arctic char. Accumulation of dioxin-like compounds of up to 8 times higher levels than the lowest observed effective level for egg mortality in temperate salmonid fish raise concern that residential Arctic char might be at risk for adverse effects at the individual and population level. In this study the aim was to compare the toxic potency of pollutants in Arctic char from the contaminated Lake Ellasjøen with those from the less contaminated Lake Laksvatn at Bear Island. This was done by in situ sampling, extraction, and fractionation of liver tissues from eleven populations. The following POP fractions (F1-2) were produced: F1- nonpolar POPs such as PCBs, PBDEs and most of the nonpolar pesticides, F2- polar pesticides and metabolites of POPs, and F3- polar POPs (phenolics such as chlorinated phenols and hydroxylated metabolites of PCBs and PBDEs). A method for isolation, cultivation and exposure of primary hepatocytes from Arctic char was observed together with the established method for primary hepatocytes from rainbow trout to investigate cytotoxic and estrogenic effects of the fractions. The estrogenic potency, measured as induction of the estrogen receptor (ER), (ER)-mediated production of vitellogenin (Vtg), was higher in liver extracts from Lake Ellasjøen than Lake Laksvatn. Although primary hepatocytes from both species displayed estrogenic activity in response to the F3 from Laksvatn fish, higher Vtg induction was observed in rainbow trout hepatocytes than in hepatocytes from Arctic char. However, the Arctic char hepatocytes were more susceptible for cytotoxic effects than rainbow trout hepatocytes. F3 and F2 from both fish populations (Lake Ellasjøen and Lake Laksvatn) appeared to have similar effect on cell viability with F3 having largest effect. Chemical analysis was performed to identify potential contributors to the observed effects. The project was funded by the Norwegian Research Council, project No. 221373.

**MO179**

Ultrasound: A novel approach to non-lethally measure hepatosomatic index in sentinel fish for environmental monitoring programs

A.R. Divergent, Wageningen University / Department of Toxicology; N. van den Berg, N. van den Brink, Wageningen University / Dept of Toxicology.

Cadmium can induce toxic effects via different mechanisms, e.g. depletion of cellular antioxidants leading to increase of ROS levels and induction of apoptosis. Such effects have been addressed in different types of tissues and cells, including the immune system. However, these mechanisms of toxicity may have differential impacts on physiological and histological functions, depending on the type of target cell. In this study, the immunomodulatory effects of cadmium between two marine immune cell models in vitro, macrophages and mast cells.

**MO181**

Divergent immunomodulatory effects of cadmium between two marine immune cell models in vitro, macrophages and mast cells.

A. Divergent, Wageningen University / Department of Toxicology; N.H. van den Berg, N. van den Brink, Wageningen University / Dept of Toxicology.

Cadmium can induce toxic effects via different mechanisms, e.g. depletion of cellular antioxidants leading to increase of ROS levels and induction of apoptosis. Such effects have been addressed in different types of tissues and cells, including the immune system. However, these mechanisms of toxicity may have differential impacts on physiological and histological functions, depending on the type of target cell. In this study, the immunomodulatory effects of cadmium between two marine immune cell models in vitro, macrophages and mast cells.

**MO182**

Changes in protein expression of primary sea turtle cells exposed to contaminants indicate the potential for in vitro proteomics as a high throughput tool to support biomarker discovery.

S.J. Chausia, Griffith University - Smart Water Research Centre / Australian Rivers Institute; F.D. Leusch, Griffith University / Australian Rivers Institute; A. Nouwens, The University of Queensland / School of Chemistry and Molecular Biology; J. van de Merwe, Griffith University / Australian Rivers Institute.
The development of biomarkers of chemical exposure and effect in threatened wildlife is challenging because traditional methods for biomarker discovery that involve in vivo testing or destructive sampling cannot be utilised. These challenges have long since been acknowledged and the development of non-destructive methods for the detection of biomarkers in wildlife has been examined for many decades. However, despite these efforts, progress in this field has been slow and the development or improvement of non-destructive biomarkers for threatened wildlife has been limited. The usefulness of cell lines along with non-targeted omics has not been well explored in this context despite the potential for these methods to greatly enhance non-destructive biomarker development. In order to assess the potential of these methods for the development of biomarkers of exposure and effect, we optimised exposure and extraction methods and aimed to investigate the effects of time and dose on global protein expression in a primary green sea turtle (Chelonia mydas) skin cells were exposed to two contaminants known to accumulate in sea turtles - a polychlorinated biphenyl (PCB153) and perfluorononic acid (PFNA). The exposure was performed over 24 or 48 hours to three environmentally relevant concentrations (1 µg/L, 0.1 µg/L, and 0.01 µg/L). Global protein expression was then measured using quantitative LC/MS resulting in over 1000 unique protein identifications. Our results show that a large number of proteins, over 700, were significantly differentially expressed by cells under exposure conditions and that time and concentration had significant effects on overall differential expression as well as on the expression of individual proteins. Most significantly, a biomarker of PCB exposure that has previously been identified in sea turtles (superoxide dismutase) was expressed by cells exposed to PCB indicating that these methods can potentially reflect biomarkers measured in whole organisms. Overall, the results from this study provide insight into the effects that time, dose and treatment have on global protein expression of green sea turtles cells, as well as preliminary evidence for the usefulness of non-targeted proteomics for biomarker discovery. This indicates the potential of the methods described here to support higher throughput confirmation of biomarkers of exposure and effect while reducing the need for invasive sampling on threatened wildlife.

MO183 Baseline vs. Reactive Toxicity toward the Nematode C. elegans as Alternative Bioassay

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MO184 Oxidative Activation of Pro-Electrophiles Mediated by an Fe-loaded Zeolite - A Nonanalomal Tool for Mimicking Phase I Metabolism

J. Moldr, Helmholtz centre for environmental research - UFZ / Ecological chemistry; A. Böhme, UFZ - Helmholtz Centre for Environmental Research / Ecological Chemistry; A. Becker, Leipzig University; G. Schuurmann, Helmholtz centre for environmental research - UFZ / Department of Ecological Chemistry

The electrophilicity of a chemical substance can have a significant impact on its toxicological profile, as this property determines the chemical's ability to form adducts with electron rich sites of proteins, lipid components, DNA and RNA. In many cases, this type of adduct formation is the molecular initiating event (MIE) of a complex adverse outcome pathway. Kinetic chemosays, which address this type of MIE, are used for the quantification of a test chemicals' reactivity towards typical biological targets, and have facilitated the development of e.g. models for the prediction of the aquatic acute toxicity or skin sensitization potential.1,2 Apart from chemicals that possess electrophilic substructures in their initial form, there are compounds that can become electrophilic only after abiotic or biotic oxidation. The reactive toxicity of these chemicals, also described as pro-electrophiles, is often difficult to characterize with simplified model systems like chemosays or in vitro bioassays because these methods typically do not sufficiently include an activation step. Our presentation introduces a new tool, based on an Fe-loaded zeolite, that is able to mediate the transformation of pro-electrophoric phenol and dihydroxybenzene derivatives. In vitro reactions are trapped by coincubation with the tripeptide WCG (tryptophan, cysteine, glycine) and analyzed using high performance liquid chromatography coupled to tandem mass spectrometry. Profiling of the obtained adduct patterns enable the identification of formed electropholes, and provides new insights into the detoxification pathways causing the reactive toxicity of pro-electrophiles. The authors gratefully acknowledge the EU-funded project OSIRIS (GOCE-CT-2007-037017) and the BMBF-funded project ProFlaTox (FKZ 031A422A and 031A422B) for financial support. [1] Böhme A, Laqua A, Schüürmann G 2016. Chem. Res. Toxicol. 29: 952-962. [2] Mulliner D, Schüürmann G 2013. Mol. Inf. 32: 98-107. [3] Chipinda I, Ajibola RO, Morakinyo MK, Ruwanna TB, Simroy MH, Siegel PD 2010. Chem. Res. Toxicol. 23: 918-925. [4] Böhme A, Thaens D, Paschke A, Schüürmann G 2009. Chem. Res. Tox. 22: 742-750.

MO185 Integrated assessment of aquatic ecotoxicity for regulatory purposes c. cappelli, IRCCS - Istituto di Ricerche Farmacologiche Mario Negri; C. Toma, IRCCS - Istituto di Ricerche Farmacologiche Mario Negri / Department of Environmental Health Sciences; A. Manganaro, Kode s.r.l.; D. Gadaleta, IRCCS - Istituto di Ricerche Farmacologiche Mario Negri / Department of Environmental Health Sciences; J. Arning, German Environment Agency UBA; A. Biegel-Engeler, German Environment Agency - UBA / Chemicals; E. Benfenati, IRCCS Istituto di Ricerche Farmacologiche Mario Negri / Department of Environmental Health Sciences

The REACH regulation requires the assessment of the CMR and PBT properties of the chemicals produced or imported in EU in amounts exceeding 10 t/a in order to protect the human health and the environment. The JANUS project aims at the development of a JAVA application to prioritize and assess the chemicals according to the PBT, CMR and endocrine disruption properties with uncertainties estimation. To assess the ecotoxicity part of the Registry, we developed six continuous QSARs for acute and chronic aquatic endpoints for the main toxic levels: EC50 96h and NOEC 96h algae (Raphidocelis subcapitata), EC50 48h and NOEC 21d Daphnia magna, LC50 96h fish (Oryzias latipes) and NOEC fish (more fish species). We used gsetool and VSURF to select the DRAGON descriptors and the tree ensemble (random forest) method to derive the models, obtaining good performance (R² up to 0.96 on the training set and up to 0.78 on the validation set when the applicability domain is considered). We used the three new QSARs on chronic endpoints to perform the assessment of chemicals; we used the three new QSARs and the seven ones implemented in the VEGA platform (https://www.vegahub.eu/) on acute endpoints for screening purposes (two QSARs for Daphnia magna, two generic QSARS for fish, three QSARs for specific fish species). The ecotoxicity workflow is divided in three parts: algae, Daphnia magna and fish. For each part and each endpoint, the workflow integrates the experimental values (if any), the QSAR predictions and their reliabilities. The experimental values have a higher reliability than the predictions. The reliability takes into account the intra- and interspecifc variability, the most sensitive species and the applicability domain index of the predictions. The users can choose a consensus or a worst-case approach. The experimental values and the predictions are used to support the regulatory thresholds to verify if the ecotoxicity criterion is fulfilled for each trophic level. The final assessment for ecotoxicity is based on the integration of the assessment of the trophic levels (the values and their reliabilities) and the number of trophic levels that fulfil the ecotoxicity criteria. The scheme will be applied to other categories of chemicals, such as the biocides within the LIFE COMBASE project. The authors thank the projects JANUS (contract Z 6 710/20 - 37165 614 0) funded project ProHapTox (FKZ 031A422A and 031A422B) for financial support. [1] Böhme A, Thaens D, Paschke A, Schüürmann G 2009. Environ. Toxicol. Chem. 28: 1700-1708, 2009. [2] Böhme A, Thaens D, Paschke A, Schüürmann G 2009. Mol. Inf. 8: 134-143. [3] Chipinda I, Ajibola RO, Morakinyo MK, Ruwanna TB, Simroy MH, Siegel PD 2010. Chem. Res. Toxicol. 23: 918-925. [4] Böhme A, Thaens D, Paschke A, Schüürmann G 2009. Chem. Res. Tox. 22: 742-750.

MO186 An integrated testing strategy to fill data gaps for environmental risk assessment of isoo-alcohols

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Environmental risk assessments require quality data to provide defensible environmental quality benchmarks. Quantitative Structure Activity Relations (QSAR) endpoint estimates are often appropriate for alcohols with a very strong environmental quality benchmarks. Quantitative Structure Activity Relations (QSAR) endpoint estimates are often appropriate for alcohols with a very strong
Aquatic toxicity testing with algae, daphnids and fathead minnows (OECD 201, ISO 20665 and OECD 210) was performed with isoocat and isoctadecan. The study objective was to employ a testing program consisting of long-term fish (limit test), invertebrate and algal toxicity tests to demonstrate that QSR estimations accurately predict aquatic effects from long-term continuous exposure to these substances, further supporting the use of QSR models across a range of isoelectrodes. The data demonstrate that the QSR model employed accurately characterized the hazard of iso-alcohols and is protective of these endtypes. Moreover, this combined information, by demonstrating a regular and predictable pattern of toxicity amongst these substances, further justifies read-across between substances for other endpoints (such as bioaccumulation) and supports efficient use of data for general purpose risk assessments.

MO187
Looking for an alternative to glyphosate-based herbicides
Vi. Lioussa, K. Eisner, S. Limbeck, D. Rünzler, University of Applied Sciences Technikum Wien / Department of Biochemical Engineering
Glyphosate-based herbicides are widely used in agriculture. When these products were originally introduced on the market, they were intended to affect only target species i.e. plants. However, over the past decades there is growing evidence on the toxicity and genotoxicity of glyphosate on non-target species. On 27th November 2017 the EU member states agreed on a five-year renewal period for the use of glyphosate-based herbicides. However, in case glyphosate-based herbicides become prohibited eventually, the availability of alternative active substances will become an urgent need. Nonanonic acid (a.k.a. pelargonic acid) is a biologically derived substance considered as an environmentally friendly herbicide. Its toxicity level to mammals is low and is also not expected to have adverse effects on non-target organisms. The aim of the present study was to compare the toxicity levels of glyphosate and a glyphosate based herbicide against pelargonic acid and a pelargonic acid-based herbicide on aquatic ecosystems using zebrafish as a model organism. In order to do so, we investigated the effect of both active substances and their formulations on the developmental stages of zebrafish embryos (OECD Guideline, Test No 236). The corresponding values of LC50 were calculated. The larvae that hatched from the acute toxicity tests were recorded in the observation chamber DanioVision and their swimming behavior was evaluated in EthoVison software. The potential effect of the tested substances on the respiratory system of aquatic organisms was investigated in vitro by performing the Neutral Red Uptake assay on the trout-derived gill cell line RTgill-W1. Results of our in vivo and in vitro tests indicate that pelargonic acid and its formulation are more toxic (acute toxicity) than glyphosate and its based-herbicide. Furthermore, the behavioral assay indicates a potential for a neurotoxic effect of pelargonic acid on zebrafish larvae. To our opinion, the results from both in vivo and in vitro assays, and in vitro assays from other research groups, will be of great interest for the future development of an environmentally friendly herbicides. In future studies we want to focus on in vivo test of pelargonic acid and other pelargonic acid-based formulations.

MO188
Chemoassay Profiling of Salicylates to Assess Their Reactive Toxicity
A. Böhme, A. Thaens, D. Paschke, A. Schüürmann, G. Helmholz Centre for Environmental Research / Ecological chemistry; G. Schuurmann, Helmholz centre for environmental research - UFZ / Department of Ecological Chemistry
Salicylates are widely used as fragrance additives or UV light absorbers in cosmetic products. For assessing the toxicological hazard of organic cosmetics and consumer care products, and thus can contribute to the human health. The BIONIC model could apply such key parameters for ionogenic surfactants. The phospholipid-water partition coefficient is considered to be the dominant contributor to the overall tissue-water partition coefficient for ionogenic surfactants, because membranes lipids allow for both ionic interactions at the zwitterionic head groups and hydrophobic interactions at the membrane core. Sorption experiments verify orders of magnitude higher affinities of ionogenic surfactants compared to surfactant or detergent. In order to do so, we investigated the effect of both active substances and their formulations on the developmental stages of zebrafish embryos (OECD Guideline, Test No 236). The corresponding values of LC50 were calculated. The larvae that hatched from the acute toxicity tests were recorded in the observation chamber DanioVision and their swimming behavior was evaluated in EthoVison software. The potential effect of the tested substances on the respiratory system of aquatic organisms was investigated in vitro by performing the Neutral Red Uptake assay on the trout-derived gill cell line RTgill-W1. Results of our in vivo and in vitro tests indicate that pelargonic acid and its formulation are more toxic (acute toxicity) than glyphosate and its based-herbicide. Furthermore, the behavioral assay indicates a potential for a neurotoxic effect of pelargonic acid on zebrafish larvae. To our opinion, the results from both in vivo and in vitro assays, and in vitro assays from other research groups, will be of great interest for the future development of an environmentally friendly herbicides. In future studies we want to focus on in vivo test of pelargonic acid and other pelargonic acid-based formulations.

MO190
The Xenopus Embryonic Thyroid Signalling Assay (XETA) for assessment of effluents contamination in thyroid active molecules,
D. Du Pasquier, Watchfrog S.A.; S. Guerin, V. Rocher, SIAAP; J. Mougel, AQUIRIS; A. Tindall, G.F. Lemkine, Watchfrog S.A.
The Xenopus Embryonic Thyroid signalling Assay (XETA) was designed as a screening test to provide information on the potential of a test substance or a sample to alter the normal functions of the thyroid system. The XETA provides a rapid (< 72h) way to measure the response of embryonic stage tadpoles to potential thyroid disrupting chemicals, allowing a efficient method for screening thyroid disruptors. XETA could provide an alternative to complex in vivo tests. It can be used for screening large number of molecules or testing environmental samples that couldn’t be stored or sampled in large quantities. OECD is currently validating this in vivo assay, the final stage of validation has been completed in 2017 and it is expected that the XETA may be approved as an OECD Test Guideline by 2019. OCDE validation focus on using the XETA to test pure chemicals but this test could be particularly useful for the hazard assessment of effluents. During the past 12 years we applied this assay to effluents including municipal wastewater, treated and untreated sewage, recycled water and rainwater. A part of our studies focused on performances of wastewater treatment plant (WWTP). Assessing the quality of the WWTPs outlets for endocrine active molecules is a major challenge for reaching the good ecological status of the natural water bodies defines by the EU Water Framework Directive. An evolution of the water treatment process is required to remove the endocrine effect present in the wastewater. The evolution should include measurement of the endocrine effect to allow the control of the treatment performance. Only Bioassays have the capacity to integrate the effect of all compounds present into a global hormonal potential and are therefore promising tools for future development of in-line assessment. Our results using the XETA on WWTP effluents showed 1) Daily variations of the thyroid effect in wastewater not linked to economic activities and rainfall 2) Most WWTP effluent still contains thyroid active molecules, results from chemical analysis performed on the same samples indicated a correlation between the total micropollutant load, a) wastewater linked to economic activities and rainfall 2) Most WWTP effluent still contains thyroid active molecules, results from chemical analysis performed on the same samples indicated a correlation between the total micropollutant load, b) wastewater linked to economic activities and rainfall 3) A minor part of the thyroid effect removal occurs during and same samples indicted a correlation between the total micropollutant load, a wastewater linked to economic activities and rainfall 2) Most WWTP effluent still contains thyroid active molecules, results from chemical analysis performed on the same samples indicated a correlation between the total micropollutant load, b wastewater linked to economic activities and rainfall 3) A minor part of the thyroid effect removal occurs during and Marketplace for all aquatic excess testing with algae, daphnids and fathead minnows (OECD 201, ISO 20665 and OECD 210) was performed with isoocat and isoctadecan. The study objective was to employ a testing program consisting of long-term fish (limit test), invertebrate and algal toxicity tests to demonstrate that QSR estimations accurately predict aquatic effects from long-term continuous exposure to these substances, further supporting the use of QSR models across a range of isoelectrodes. The data demonstrate that the QSR model employed accurately characterized the hazard of iso-alcohols and is protective of these endtypes. Moreover, this combined information, by demonstrating a regular and predictable pattern of toxicity amongst these substances, further justifies read-across between substances for other endpoints (such as bioaccumulation) and supports efficient use of data for general purpose risk assessments.

MO191
Advances on locomotion detection of Daphnia magna, Artemia franciscana and Parameca caudatum
E.M. Salzer, V. Lioussa, X. Monforte Dila, R. Rünzler, University of Applied Sciences Technikum Wien / Department of Biochemical Engineering
Animal behavior is complex and multidimensional. Over the past decades researchers tried to qualify and quantify it, in order to understand and predict it. The advances in this field are so radical that they actually formed a new scientific domain called “computational ethology”. A major gap in this field is that most
MO194

In silico site-directed mutagenesis informs species-specific predictions of chemical susceptibility derived from the Sequence Alignment to Predict Across Species Susceptibility (SeqAPASS) tool

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There are some concerns for environmental impacts of the pharmaceuticals due to the unintended environmental effects, which may be different from biological medicinal effects. Therefore, medical regulatory agencies require the assessment reports of environmental impacts by new drugs before marketing. It would be useful to predict the ecotoxicity of the new drug at the developmental stage, because the ecotoxicology studies are usually conducted at the final drug developmental stage just before submission. To validate the current in silico prediction tool, we evaluated the prediction for ecotoxicity prediction by the ECOSAR software, with high sensitivity for evaluation of industrial chemicals. In the last year, we evaluated the prediction performance of Daphnia magna reproduction and of the fish chronic toxicity. In this study, we evaluated prediction performance of the acute and chronic toxicity for algae. We used the ecotoxicity test data sets of about 100 pharmaceuticals. The EC50 values for the acute toxicity and the ChV for the chronic toxicity were compared with the prediction values estimated by the ECOSAR. The percentages of the pharmaceuticals of which the predictive values are different in less than one digit from the actual measured values are 43% and 44% for acute and for chronic toxicity, respectively. Overall applicability of toxicity prediction for algae was similar to that of toxicity for Daphnia magna or fish. In the case of Daphnia magna or fish, some antibiotics, anti-cancer, central nervous system agents with lower LogPow were underestimated. However, there is little dependency of LogPow in the case of Algae toxicity. Most of chemicals with aliphatic amines had tendency to be underestimated. This difference may reflect on the mode of actions between Daphnia/fish and Algae. In order to improve the predictability of the in silico ecotoxicity QSAR tool, more researches on discovering the structure dependent toxicological profile of Algae would be needed. This work was supported by the Research on Regulatory Science of Pharmaceuticals and Medical Devices from Japan Agency for Medical Research and Development, AMED.

MO195

SeqAPASS to Evaluate Conservation of High-Throughput Screening Targets Across Taxa


Cell-based high-throughput screening (HTS) and computational technologies are being applied as tools for toxicity testing in the 21st century. The U.S. Environmental Protection Agency (EPA) embraced these technologies and created the ToxCast Program in 2007, which has served as a screening and prioritization tool for thousands of chemicals. The rapid and automated screening methods take advantage of hundreds of (primarily) mammalian-based HTS assays for identifying biological activity suggestive of potential toxic effects. The data can aid in identifying chemicals that are most likely to impact biological pathways that lead to adverse health effects. To realize the full potential of the ToxCast data for predicting adverse effects to both humans and wildlife, it is necessary to understand how broadly these data may plausibly be extrapolated across species. Therefore, the U.S. EPA Sequence Alignment to Predict Across Species Susceptibility (SeqAPASS) tool was used to evaluate conservation of the 460 protein targets represented in the ToxCast assay suite. The SeqAPASS query sequence was selected based on the model organism used in the ToxCast assay (e.g., human, cattle, chimpanzee, guinea pig, rabbit, rat, mouse, pig, or sheep). Similarity of primary amino acid sequences and sequences from appropriate functional domains were compared across species to establish a relative indicator for a wide range of environmental contaminants. Furthermore, changes in their locomotion could be used as an endpoint when studying neurotoxic effects. As a result, the aim of the present study was to improve the current tracking techniques of Daphnia magna, Artemia franciscana and Paramecium caudatum. In order to do so, custom-made, portable multi-well plates were designed and fabricated or swimming behavior can be easily observed and measured. The horizontal and/or vertical tracking of the tested species were performed with the software EthoVision. The results of the present study showed that our custom-made plates had a higher tracking efficiency and a higher reproducibility score compared to the commercially available multi-well plates. Therefore, the custom-made plates are a more efficient and cost efficient plates can be implemented on behavioral and ecotoxicological studies on small-sized aquatic invertebrates and planktonic organisms in any lab with an access to a tracking system. Financial support from the City of Vienna project ÖkoToxologie (MA 2.5 - Project 15-06) is gratefully acknowledged.

MO196

Survival and Teratogenic Evaluation of 91 compounds with environmental impact

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ZeClinics (www.zeclinics.com) is a biotech company interested in developing efficient and reliable zebrafish screening tests to predict compound toxicity. In recent years, the rapid growth in organ replacement and 3D printing of artificial organs is contributing to define a universal set of rules – incubation time, chorion/no chorion, analysis timing, type of end points, analysis procedure, etc. – that can be applied by all the zebrafish toxicology community (SOP like protocols) and, eventually, to become the base for applying towards regulatory approval for the standardized test. In this study, we have performed a Developmental Toxicity Test on the NTP 91 compound list. For each compound, 20 zebrafish embryos were exposed to the test compound at concentrations (Log3 dose/response curve: 100 µM, 33 µM, 10 µM, 3.3 µM, and 1 µM) for a single biological replicate. Experiments were performed in chorionated embryos from 3 hpf to 96 hpf. Endpoints were analysed at 24, 48 and 96 hpf. Quantified phenotypes include mortality rate and teratogenic endpoints such as body deformity, scoliosis, pigmentation, heart edema and motor behaviour. It is important to note that the majority of the provided compounds were already dissolved in DMSO at 10 mM. This fact limited the range of maximum concentrations tested, which might have impacted in a lower-than-expected correlation between zebrafish and human data. In fact, 49/91 compounds did not
show any toxic phenotype at the maximum evaluated concentration. On the other hand, 39 displayed mortality and teratogenic phenotypes. Among them, the most toxic compounds were Saytex CP-2000, 4,4'-hexafluoroisopropylidene diphenol, 3-lodo-2-propynyl n-butylcarbamate, diethylishtribestrol, hexachlorophene, methylmercury chloride, rotenone and tetraethylthiram disulfide.

MO196

MPA - an alternative for the standard procedure of Ames Test
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The Salmonella/microsome assay (Ames Test) is the most widely used mutagenicity test for evaluation of pure chemicals and environmental samples. There are several protocols available in the literature, including those that reduce the amount of sample needed for testing with liquid and agar media. There is a miniaturized version using liquid media called Microplate Fluctuation Protocol (MFP) that has been extensively used specially in Europe. It is has similar sensitivity with the standard Ames as well as other protocols and good performance in interlaboratory studies. However, the MFP has some disadvantages such as being difficult to apply with strains with low and high spontaneous mutation frequencies. Another miniaturized version of the Ames test is the microsuspension assay, which is 13 to 20 times more sensitive than the standard protocol. It is performed 5X concentrated bacteria and less sample and S9 mixture but still uses conventional petri dishes (90 x 15 mm). It has been extensively used for environmental samples testing, including in Effect Directed Analysis (EDA). The objective of this study was to miniaturize of the microsuspension Salmonella/microsome assay using agar microplates under the concept of the 3R principle. The conventional plates were replaced by plates with 12 micro wells. For validation of this miniaturization, we selected 13 known more or less potent mutagenic compounds. Six were tested only without metabolic activation (S9) and the other 7 were tested only with S9 using three Salmonella tester strains that were selected based on their different spontaneous reversion frequencies (low, mean and high). The miniaturization procedure conditions were made as similar as possible to the Microsuspension protocol, using the same testing design, metabolic activation, and data interpretation. Each test was conducted in parallel. MPA and Microsuspension protocols showed 100% agreement, qualitatively and quantitatively. MPA is less laborious, uses less sample, materials, and reagents reducing overall costs. The amount of sample required for testing is at less 20 times less in comparison with the standard Ames assay. We conclude that MPA is a promising tool and could be used in substitution of the standard Ames procedure especially in situations where sample quantity is a limitation such as impurities, drugs in development and environmental monitoring studies.

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SETAC Animal Alternatives Interest Group
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Bioavailability and realistic risk assessment of organic chemicals (P)

MO198

The necessity of OASIS bead and polyethersulfone membrane extraction for the Polar Organic Chemical Integrative Samplers (POCIS) calibration: a case study for alkylphenol monitoring in produced water
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Produced water (PW) is one of the largest discharges from the oil and gas industry and includes formation and injected water. It contains several toxic compounds such as polycyclic aromatic hydrocarbons (PAHs), alkylphenols (APs), heavy metals, etc. PW is usually treated on the platform and then directly discharged into the sea. Despite the low content of toxic compounds that remain in PW after treatment, the large volume of PW lead to high total amounts of toxic compounds discharged every year into the sea; thus an effective tool for monitoring the hydrophilic organic compounds (HpOCs) is necessary. Passive sampler devices (PSDs) are the most common tools for monitoring a wide range of HpOCs, such as polycyclic aromatic hydrocarbons (PAHs), alkylphenols (APs). Polar organic chemicals (HOCs) is measured in seawater using a wide range of HpOCs. It allows the time weighted average (TWA) concentration to be measured and thus assess fluctuation in discharge concentrations. POCIS is composed of a sorbent (OASIS beads), two polyethersulfone (PES) membranes and two stainless steel rings. POCIS is calibrated by evaluating the sampling rate (R), which is correlated with the contaminant concentration in the water and in the sampler, and is usually assessed by extracting the OASIS beads alone. We evaluated the effect of the PES membranes on AP uptake and, for the first time, calculated the R, following the extraction of both the sorbent and the PES membranes. The study demonstrated that there was a lag phase in uptake for APs, and that APs with log Kow>5 were accumulated more efficiently in the PES membranes. The extraction of both the PES membranes and the OASIS beads is thus needed when working with the POCIS in order to capture low contaminant concentrations and allow the detection of the less hydrophobic APs. This can be very useful in environmental applications because it may justify the use of only one passive sampler to monitor a wider range of contaminants.

MO199

In situ passive sampling methods to measure freely dissolved concentration of PAHs in contaminated soil: comparison with ex situ measurements and evaluation over one year
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Bioavailability studies can be used to improve risk assessment and legislation relating to soil and sediment PAH concentrations in the pore water of soils under field conditions (HOC). Over the past decade, researchers have successfully developed various passive sampling (PS) methods to assess the freely dissolved concentrations of HOC in soil pore water or suspensions (CPOC). The CPOC play a key role for environmental fate and toxic effects of these compounds. Field conditions such as temperature, ionic strength or soil water content may influence the distribution of HOCs. PAHs are accounted for by the sum of a free and a bound fraction (in situ PS method) or by providing promising results to measure CPOC in the pore water of sediments, there is still very little information on the suitability of these methods for their application to soils, particularly under unsaturated water conditions. Here, we present the results of in situ PS concentrations of polycyclic aromatic hydrocarbons (PAHs) in situ compared with laboratory soil field studies. The soils were located in peat bogs and hence permanently water-saturated, while the other three were located in grassland and thus not saturated. Low density polyethylene (LDPE) was used as PS method. The samplers were deployed in situ covering a depth of 20 cm below the soil surface. Concentrations were assessed at all sites after six, nine and twelve months of exposure. For comparison, soil samples from the same locations were also evaluated using a conventional ex situ soil suspension method. The main objectives of this study were (1) to compare the measurements obtained with the two sampling methods, (2) to assess the influence of soil water saturation of the measurements (unsaturated versus saturated), and (3) to determine the role of seasonal variation (temperature and precipitation variation) and exposure time on the results of the in situ PS method. To our knowledge, this is the first experiment where PS methods were used to determine PAH concentrations in the pore water of soils under field conditions in situ and to study the impact of soil water saturation. This study will help to find out whether in situ PS methods in soils are a tool to be potentially included in risk assessment and legislation.

MO200

Bioaccumulation of native and spiked p,p‘-DDE by Eisenia andrei in γ-stabilized and non-stabilized soils
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The fate of organic chemicals and their metabolites in soils is often investigated in model matrices having undergone various pretreatment steps that may qualitatively or quantitatively disturb results. Presently, effects associated to γ-irradiation, spiking and dwelling of earthworms were studied in field-cased soils: (sterilization after contamination) and freshly spiked (sterilization prior to contamination) soils for the case of 1,1-dichloro-2,2-bis(p-chlorophenyl) etylene (p,p‘-DDE). Changes in its sorption and bioavailability were linked to the changes in soil organic matter (SOM) chemistry measured by Diffuse Reflectance Infrared Fourier Transform (DRIFT) spectroscopy. Qualitative interpretation of obtained DRIFT spectra revealed SOM chemical changes in a reduction of relative intensities of aliphatic moieties (sterilization), in bands of hydroxyl, aromatic, and aliphatic moieties (spiking), and of reduction in bands of aromatic components accompanied by an increase of aliphatic moieties (earthworms dwelling). Using DRIFT, changes induced by spiking and earthworms were noted to be more pronounced compared to sterilization. The variation of bioaccumulation factors (BFR) of native and spiked p,p‘-DDE in sterile and non-sterile soils was limited to a factor of 1.5, depending on the incubation time and the particular approach used for BAF calculation. Despite the absence of quantitative effects of γ-irradiation on p,p‘-DDE bioaccumulation, the uptake kinetics were shown to vary between
non-stereile and sterile soils. Sterilization appeared to increase uptake rates and reduce the influence of p,p'-DDE-soil contact time on bioaccumulation. These effects might be attributed to the effects of γ-irradiation on SOM chemistry alone or in combination with earthworms. Following our findings, γ-irradiation can be recommended as a relatively non-destructive method that is not expected to significantly affect risk assessment of bioaccumulative chemicals. However, in mechanistic studies the possible side-effects brought about by γ-irradiation should be taken into consideration.

MO201 Dissipation in soil and bioavailability to earthworms of two fungicides: comparison of laboratory and field experiments
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The pesticide fate in soil is currently studied through laboratory experiments, using homogenized soil and controlled incubation conditions. However, the representativeness of such experiments is questionable regarding field conditions, i.e., soil heterogeneity, vegetation cover and climatic variations. Upon the few studies performed simultaneously under field and laboratory, the pesticides persistence and/or sorption on soil were evaluated as different; thus bioavailability could also be different. This study aimed at comparing the dissipation and the bioavailability of dimoxystrobin and epoxiconazole (two fungicides used in the commercial formulation of Swing Gold®) in a loamy soil and in earthworms under laboratory and field conditions. Field experiments were conducted in a meadow located in Versailles (France) on 100 m² plots. The field soil and earthworms were regularly sampled after the treatment, over one year (April to April). Laboratory experiments were performed by mixing the fungicide solution with surface soil issuing from the same field, and incubated in dark, at 15°C and constant humidity. In both cases, four replicates were performed using the recommended Swing Gold® dose, extra doses adapted to observe ecotoxicological effects and controls. The dimoxystrobin and epoxiconazole concentrations in soil were determined by an exhaustive extraction method and, to evaluate their availability, with a mild method engaging hydroxypropyl-β-cyclodextrin. At the same time, the bioavailability of the two fungicides was evaluated by determining their concentrations in exposed earthworms Aporrectodea icterica and Aporrectodea caliginosa. All analyses were performed by UHPLC-MS/MS. Under field conditions and five days after pesticide application, only 10% to 45% of pesticide residues were measured in topsoil, with high heterogeneity between replicates. At one month, the concentrations in soil increased, probably due to a plant-soil transfer. By contrast, applied dose was observed at initial time under laboratory conditions. For later dates and in both cases, dissipation was observed. The available fraction showed homogeneous rates under laboratory conditions, and highly heterogeneous ones in the field. However, the ratio of available/total concentrations showed the same trend of fate for the two tested substances. The bioavailability of the two fungicides was also different between field and laboratory conditions in terms of heterogeneity.

MO202 Experimental assessment of specific plant uptake factor of 1,2,4-triazole with different concentrations in wheat
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Environmetally significant risk assessment decisions in EU for Plant Protection Products (PPP) regulation are based on information obtained from Environmental Fate models. Such models (e.g., FOCUS PEARL and PELMOT) attempt to quantify the potential for movement of pesticides and their metabolites to ground water and require data derived from experimental studies based on validated guidelines. Currently, guidelines detailing degradation and absorption processes of pesticides and their metabolites in soil do not accurately account for uptake by plants via root system, with plant uptake compartments only being given a default value. As plant uptake affects the environmental availability of these substances, accurately quantifying this effect is a critical result in the risk assessment predictions. The plant uptake describes the process of translocation of dissolved compounds in the soil pore water to the plant via the transpiration stream and it can be described using the plant uptake factor (PUF) – uptake into shoots and roots – or the transpiration stream concentration factor (TSCF) – uptake into shoots. Recent work by the ECPA/IVA Working Group “Plant Uptake Factor” has produced a draft working protocol designed to experimentally determine the uptake of active substances as well as metabolites via plant roots. The purpose of the present study was to obtain reliable substance-specific plant uptake data – with different root zone exposure concentrations – using the study design proposed in draft working protocol. The set-up of the experiment was chosen to enable optimal growth of the test plants – wheat seedlings – grown in a hydroponic system under controlled environmental conditions. At BBCH 13 (3 leaves unfolded) “4-chloro labelled 1,2,4-triazole was spiked into the hydroponic solution at different concentrations and the plant root system was exposed for 8 days. Mass balance – calculated from the sum of radioactivity found in the hydroponic solution, root wash plus roots and shoot tissue – and transpiration – calculated gravimetrically – were determined. The experimental data obtained were used to calculate uptake parameters – PUF and TSCF – according to the formulas mentioned in the literature.

MO203 LFER Models for Partition Coefficients of Environmental Concern
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Traditionally, partition coefficients of environmental concern, including bioconcentration factors (BCF) and soil or sediment sorption coefficients, are predicted using approaches based on simple linear relationships with the octanol/water partition coefficient (Kow). Recently, more sophisticated prediction models have been developed and applied, including LFER approaches. Such approaches allow distinguishing between separate uptake mechanisms and, however, this requires data on distinct partitioning processes, which are rather scarce. For theoretical models without the need for additional experiments, the partition coefficients for these separate processes have to be estimated. While these coefficients basically could be roughly estimated from Kow, the more sophisticated LFER equations, known as Abraham models, are preferred for such predictions. The and the respective partition coefficients are PBS, 18.5%, 18.1%, 11.1%, 1.0% and 0.3%, respectively. Two-dimensional partition coefficients of neutral organic compounds between water and immobilised artificial membranes (IAM), liposomes (membrane lipids), triolein (storage lipids), and sediment. The development of the models was based on new experimental data, as determined by the authors. Possible sources of the required Abraham parameters are examined, compared and discussed. Particular attention is given to the applicability domain of the models. Acknowledgment: This study was financially supported by the European Union 7th Framework Programme SOLUTIONS (FP7–ENV-2013) of the under grant agreement no. 603437.

MO204 Influence of grain size on the bioavailability and bioaccumulation of sediment-associated cypermethrin to benthic invertebrates
H. Li, J. You, Jinan University / School of Environment Sediment particle-size distribution is an important factor influencing the bioavailability and toxicity of hydrophobic organic contaminants (HOCs) in sediment. Cypermethrin, a pyrethroid was used as an example in the current study to investigate the effect of particle size on the desorption kinetics of cypermethrin bioaccumulation potential of sediment-associated HOCs. Bioaccumulation test with oligochaete Lumbricus variegatus and two chemical techniques, namely Tenax extraction and matrix-solid phase microextraction (SPME) were applied in the current study. A field sediment was collected and wet sieved to obtain five particle-size fractions: i.e., < 20 μm, 20–63 μm, 63–180 μm, 180-500 μm and > 500 μm. The respective ratios of the five size fractions were 81.2%, 10.9%, 1.8%, 14.7%, 2.86% and 0.31%. The results of sediment characterization showed that the material, composition, surface area and adsorption capacity were significant different among sediments with different particle size, and adsorption capacity increased with decreasing particle size. In addition, the desorption rates of cypermethrin measured by Tenax extraction decreased with decreasing particle size and supported the finding that HOCs bioaccumulation potential of sediment particles than coarse sediment. The different desorption rates of cypermethrin in different particle-size sediments may influence the freely dissolved concentrations in sediment porewater, and subsequently bioaccumulation potential and toxicity.

MO205 Effect of suspended particle on polycyclic aromatic hydrocarbon (PAH) bioaccumulation by zebrafish (Danio rerio)
Y. Zhai, X. Xia, School of Environment, Beijing Normal University; X. Xiong, School of Environment, Beijing Normal University / School of Environment; L. X. x. guo, School of Environment, Beijing Normal University Hydrophobic organic compounds (HOCs) tend to be associated with suspended particles than coarse sediment. The different desorption rates of cypermethrin in different particle-size sediments may influence the freely dissolved concentrations in sediment porewater, and subsequently bioaccumulation potential and toxicity.
process. Suspended particles promoted the uptake and elimination rate constants of PAHs to zebrafish body excluding head and digestive tracts. The uptake rate constants with 0.5 g/L-suspended particle were approximately twice that without suspended particles, and the body burden in zebrafish increased by 16.4% - 109.3% for pyrene and 21.8% - 490.4% for fluoranthene during the first 8-d exposure. The findings from this study indicate that PAHs on suspended particles are partly bioavailable to zebrafish and particle ingestion is an important route in PAH biaccumulation. Therefore, it is important to consider the bioavailability of HOCs on suspended particles to improve ecological risk assessment.

MO206 Methods for Deriving Site-Specific Relative Bioavailability Factors from Agricultural Bioavailability Data

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The U.S. Department of Defense is responsible for the environmental restoration of properties that were formerly used for firearm training ranges. Re encourants of spent shell casings are determined to be a source of polycyclic aromatic hydrocarbons (PAHs) at these sites based on results from surface soil analyses and historical information demonstrating that shell targets were commonly prepared using coal tar pitch as a binding agent. It was hypothesized that the nature of the coal tar pitch/limestone mix of the shell fragments reduces the oral bioavailability of PAHs compared to that seen in animal studies using pure benzo(a)pyrene in solvents added to diets. To test this hypothesis, soil samples were collected from two sites to provide a range of PAH concentrations. Female B6C3F1 mice were fed diets amended with soil or soil extracts at a ratio of 5% in the diet for fourteen days. For benzo(a)pyrene (BaP), the fraction of total dose excreted in the urine (FUE) was determined for the soil- and soil extract-amended treatment groups. The Relative Bioavailability Factor (RBAF) is the ratio of the FUE in animals treated with soil over the FUEs treated with extracts of soil. Because each soil sample was tested in four cages of mice (two for soil and two for soil extract), there are different ways of computing the RBAF of each soil and the grand RBAF for the site. Pairwise RBAFs can be determined and averaged, but the more robust way to determine a site-wide RBFA from multiple sample points is to determine a linear regression of the metabolite excretion rates versus daily dosing rates. The FUEs produced coefficients of determination (r2) that were greater than 0.83 and typically greater than 0.95, showing that the rate of BaP+ metabolite excretion was directly proportional to the daily dose rate of BaP. RBAFs were determined using Monte Carlo simulations to calculate the 95% upper confidence limit on the ratio of the soil and soil extract FUEs. The site-wide RBAF was equal to 14% for BaP. Pairwise RBAs will be compared to the RBAF resulting from the regression approach, and the regulatory precedent for the regression approach will be presented.

MO207 Accurate determination of adsorption coefficients for low adsorbing compounds - from experiment to result evaluation

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The determination of adsorption coefficients is a critical key parameter for the assessment of the leaching properties of low adsorbing compounds’ through the vertical soil profile. In case of expected low adsorption of a chemical compound, several experimental preconditions need to be considered to enable accurate adsorption parameter determinations: (1) An initial soil/solution ratio of 1/1 and (2) the liquid phase needs to be removed as completely as possible from the soil phase upon completion of the equilibrium. An experimental approach was developed and optimized allowing the efficient separation of the soil and liquid phase by centrifugation through the soil itself and a filter/frit system. Deter-

MO208 Evaluation of the swimming behavior and tactic response to atrazine of the Pseudomonas sp. strain ADP

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Atrazine, a common herbicide used to control grassy and broadleaf weeds in sugarcane, wheat, conifers, sorghum and a variety of other crops. Although since 1992 in European Union countries it has been banned owing to its toxicological effects, it remains one of the most consumed worldwide pesticide with annual consumption of about 70,000-90,000 tons. Atrazine removal from the environment depends on abiotic (photolysis and hydrolysis) and above all biotic degradation. The latter can be significantly affected by the herbivore bioavailability. The behavioral reactions of bacteria are rarely included in the biological assessment of contaminants ecotoxicity. For this reason, we investigated the swimming behaviour and tactic response of the motile atrazine-mineralizing bacterium Pseudomonas sp. strain ADP to different concentrations of the herbicide in a laboratory experiment. The tactic response was assessed by a chemical-in-capillarity method and an inverted capillary assay for the repellent reaction in association with microscopic observations. The swimming behaviour was evaluated by a computer motion analysis software (CellTrack). We observed attraction reactions at relatively high concentrations of the chemical, including at water-saturating concentrations. We also noticed that atrazine can elicit a negative tactic response at low concentrations. We also observed that the swimming patterns of Pseudomonas sp. strain ADP was affected by 28 days. The data was non-parametric, and we increased modeling by logistic. The physiological relevance of the chemioattraction to pollutants lies in the fact that these compounds serve as carbon and energy sources. The overall results suggest that we can use the behavioral responses of motile bacteria as a useful method to estimate pollutant toxicity at ecologically relevant concentrations. The bacteria behavioral assay on pollutants can be an alternative or complementary method to the current ones, because it is a high sensitivity and visualization method.

MO209 The influence of biochar on the toxic effects of imidacloprid to the lifecycle parameters of Eisenia fetida

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Imidacloprid is an organic active ingredient for various insecticides used to kill a number of biting and sucking insect pests. As a soil amendment, it can enhance sorption and reduce the bioavailability of organic contaminants. Earthworms are important soil promoters and provide various benefits to plants and they are too sensitive to various pollutants, thus they are relevant indicators of environmental change. The current study aimed to assess if biochar has the ability to decrease the effects of the toxicity of the agro-icide imidacloprid (IMID) to earthworms. Cocoons laid by earthworms that had never been in contact with any toxicant were exposed to biochar-amended and non-amended aqueous solutions of IMID for a period of 28 days. The data was non-parametric, and it increased using logit. Kruskal-Wallis ANOVA followed by Dunns’ test. The level of significance was set to p < 0.05. The results revealed that the effects of IMID on hatching success were only significant at the highest concentration of 400 mg/L. These findings indicate that the recommended field concentration of IMID (400 mg/L) has the potential to prevent cocoons of earthworms such E. fetida from hatching. This suggests that IMID could lead to decrease population growth rate or complete population collapse in such invertebrates.

MO210 Chlordecone elimination kinetics in ewes

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Chlordecone (CLD) is an organochlorine pesticide used from 1972 to 1993 against banana weevil and green banana weevil (Sitophilus oryzae and S. zeamais), respectively. CLD is persistent in the soils (concentrations are above 1 mg kg⁻¹ of dry matter). Consequently, animals can be directly contaminated by involuntary soil ingestion. Previous studies showed a CLD absorption of 100% in goats and its metabolization in humans, gerbils and pigs CLD is reduced into chlordecol (CLDOH). Then CLD was characterized in ewes (linearity of the black banana weevil (Cosmopolites sordidus) of the Skeet fragments reduces the oral bioavailability of CLD. Consequently, animals can be directly contaminated by involuntary soil ingestion. The physiological relevance of the chemioattraction to pollutants lies in the fact that these compounds serve as carbon and energy sources. The overall results suggest that we can use the behavioral responses of motile bacteria as a useful method to estimate pollutant toxicity at ecologically relevant concentrations. The bacteria behavioral assay on pollutants can be an alternative or complementary method to the current ones, because it is a high sensitivity and visualization method.

Wallis ANOVA followed by Dunns’ test. The level of significance was set to p < 0.05. The results revealed that the effects of IMID on hatching success were only significant at the highest concentration of 400 mg/L. These findings indicate that the recommended field concentration of IMID (400 mg/L) has the potential to prevent cocoons of earthworms such E. fetida from hatching. This suggests that IMID could lead to decrease population growth rate or complete population collapse in such invertebrates.
values were not significantly different (P > 0.05). Thus, it was possible to conclude that CLD toxicokinetic of CLD in ewe is linear. In urines, CLD and conjugated CLD were not quantified. By comparing the two way of CLD excretion, feces appears to be the principal route of CLD elimination. Almost 60% of the administered dose was found in feces and only 2% was found in urines. To conclude, the elimination of CLD in serum of ewe is dose-dependent with the dose. In consequence, the different results obtained for CLD in urine can be extrapolated for different levels of exposure in the range of 0-1 mg/kg BW. This study reveals the CLD metabolism in ewes which was never shown before. The principal route of CLD elimination is via the feces. These results clearly indicate the CLD elimination kinematic in ruminants and will help to decontaminate exposed animals in the French West Indies.

MO211 Development and validation of QuEChERS extraction methods with or without enzymatic pretreatment to analyze chlordene and its metabolites by HPLC-MS/MS in urine and feces of ewes

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Chlordene (CLD) is an organochlorine pesticide used from 1972 to 1993 in the French West Indies to fight against the banana black weevil. It is known to increase the risk of prostate cancer and affect development of the foetus and after birth. Nowadays, this pesticide is no longer used, because of its high persistence, it still remains in soil. Consequently, farm animals can be contaminated by soil ingestion and this is key issue for French West Indies breeding. Thus, this work tempted to collect data about the CLD elimination in ewe so as to propose a decontamination strategy. CLD is mainly eliminated in feces and low amounts of CLD can be found in urines. CLD can be metabolized into chlordecol (CLDOH) in humans, pigs and gerbils livers. Then CLD and CLDOH can be conjugated by the glucuronyltransferase. In feces, CLD was found but no conjugated metabolites were present. In urines, no conjugated metabolites were found although the authors thought they would. Actually, no information about the CLD elimination in ewe is available. These findings results were based on a former extraction method developed in 1980 to analyze CLD and its metabolites in urines and feces. The extraction was performed by liquid-liquid extraction. Quantification of the conjugated metabolites was calculated by difference between a sample with and without enzymatic treatment. The analysis was then performed by gas chromatography hyphenated with mass spectrometry. In order to have an update and more sensitive method, a new development was carried out with this work. The extraction was based on the QuEChERS methodology which is more and more used in the pesticides field. As no conjugated standards were available an update enzymatic pretreatment was set up. The analysis method was performed by liquid chromatography with tandem mass spectrometry using isotopic dilution given a reliable method. The methods were then validated according to the French standard NF V03-110 and the European Union guidelines. At the outcome of the method development, CLD and CLDOH were present in ewe feces. In urines, CLD and conjugated CLD were quantified. These results highlighted a better sensitivity of the new method and allow proving the CLD metabolism in ruminants which was never made before.

MO212 Organic Contaminants in High Mountain Areas: Where and When to find them??

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Semivolatile organic contaminants (SOC) are well known to undergo atmospheric long-range transport and enrich in remote high mountain ecosystems. To predict the risk for high mountain ecosystems it is necessary to have knowledge of the present contamination. One important aspect is their bioavailability which is more and more used for the prediction in the pesticides field. As no conjugated standards were available an update enzymatic pretreatment was set up. The analysis method was performed by liquid chromatography with tandem mass spectrometry using isotopic dilution given a reliable method. The methods were then validated according to the French standard NF V03-110 and the European Union guidelines. At the outcome of the method development, CLD and CLDOH were present in ewe feces. In urines, CLD and conjugated CLD were quantified. These results highlighted a better sensitivity of the new method and allow proving the CLD metabolism in ruminants which was never made before.

MO212 Pesticide occurrence in different apicultural matrices (honey bees, wax and pollen)

P. Calatayud-Vernich, M. Andrés Costa, Universitat de Valencia / Environmental and Food Safety Research Group; F. Calatalayud, E. Simó, Agrupación de Defensa Sanitaria Apicola (apisADs); Y. Pico, University of Valencia / Medicine Preventive Sprayed crops with pesticides are visited by honey bees during pollen and nectar collecting process. Pesticides are transported inside the hive, where both, agrochemicals from agriculture and compounds used in-ive against varroosis by beekeepers are accumulated in wax, pollen and honey bees. Samples of honey bees (45), for (43) and pollen (45) were obtained from 45 different apiaries located in Spain. The samples were extracted by a slightly modified QuEChERS procedure depending on the matrix, and then screened for 58 pesticides and its degradation products by liquid chromatography mass spectrometry (LC-MS/MS). The target analytes were chosen based on their potential toxicity to honey bees and their widespread use in plant protection or in the bee hive against varroa mite. Wax and pollen were the most contaminated matrices and exhibited a wide contamination by pyrethroids and organophosphates. Beeswax lipophilic nature and its lowest replacement rate in the hive are responsible of its highest pesticide content. Acaricides used in beekeeping such as coumaphos, chlorfenavinphos, amitraz and fluvalinate were the most frequently detected pesticides in wax. Some pesticides used in crops as organophosphate chloropyrines were detected in lower frequencies and concentrations. Pollen contamination pattern was similar to wax matrices. Acaricides applied in beekeeping were the most frequent and with the highest concentrations. Neonicotinoid acetamiprid and organophosphates chloropyrines and dimethoate were detected in pollen samples. Both insecticides are sprayed in crops and deposited on the pollen grains, which are transported to the hive during the foraging activity of the honey bees. Honey bee samples were less contaminated, although some acetamiprid was found. These results revealed the importance of the concentrations detected in the matrices analyzed, honey bee colonies health could be compromised. Assessing pesticides content in these three different apicultural matrices at the same time is a useful tool to understand the magnitude of honey colonies exposure to toxic compounds, which is one of the main causes of the progressive decline in honey bee colonies around the world.

MO214 Adaptation requirements for the use of measured BCF for a realistic risk assessment of organic chemicals.

N. Pucheux, INERIS, S. ANDRES, INERIS / Toxicological Ecotoxicological Assessment of chemical Substances

One of the main factor in the secondary poisoning risk assessment is the bioavailability of potentially hazardous organic chemicals, especially in the case of soil contaminated with persistent organic pollutants. In the context of the TROPHé project, the transfer of PCBs and PCDD/Fs to plants and invertebrates has been studied. BCF in several plants and in earthworms had been measured and different models have been calculated to predict bioconcentrations. One of the most critical aspects is the choice of the matrix that contains the contaminant. The BCF, relatable to aquatic predators can be calculated in taking in account the quantity of soil contained in the earthworms guts and the contaminant fraction measured in its fluid. According to the literature, CLD and CLDOH were present in ewe feces. In urines, CLD and conjugated CLD were quantified. These results highlighted a better sensitivity of the new method and allow proving the CLD metabolism in ruminants which was never made before.

MO215 Assessing risks from PBT substances in surface waters: possible alternatives to biota monitoring?


PCBs, polychlorinated dibenzo-p-dioxins (PCDDs), and dibenzofurans (PCDFs) are persistent organic pollutants (POT, referred to as PBTs) that are toxic to aquatic life and may bioaccumulate in fish. The exposure of aquatic organisms to PBTs is usually assessed by measuring chemical levels in biota. The Water Framework Directive (WFD) requires water bodies to be at ‘good chemical status’ by meeting Environmental Quality Standards (EQSs). Normally, chemical concentrations in water but in some cases expressed as critical concentrations in the flesh of aquatic biota (biota EQSs) have been developed for some chemicals that are persistent, bioaccumulative and toxic (PBT) with the aim of protecting predators and humans from chemical exposure via the foodchain. Biota standards are now set for 11 PBT substances or groups of substances, requiring Members States to set up monitoring regimes to assess the risks to surface waters. Biota monitoring (fish or invertebrates, depending on the substance of interest) is the most relevant sampling matrix but it is destructive, and suitable biota cannot always be found where sampling is required. As a result, the coverage offered by biota sampling programmes is much less extensive than
MO216 Risk Associated with Alternative Cleaning Method for Carrot
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ABSTRACT Risk Associated with Alternative Cleaning Method for Carrot
Introduction Carrot is a nutritional root vegetable which is loaded with beta carotene, a precursor of vitamin A. It is necessary to wash carrots in order to remove soil and other foreign materials before eating either raw or in processed form (Moos et al., 2002). It is common practice nowadays to soak carrot in detergent solution before washing to achieve better cleaning. Some components of detergent are toxic (HERA, 2013; Chuku et al., 2015). The aim of this study is to evaluate the detergent residue accumulated in carrot exposed to detergent. Methodology The first stage involved distribution of questionnaires to determine the popularity of the use of this chemical substance in washing carrot before selling to consumers. The second stage involved soaking 2 kg of fresh carrots in five increasing concentrations of Detergents 1 and Detergent 2. The carrots were soaked for 20, 40 and 60 minutes, after which they were grounded and analyzed using the titrimetric method described by IPAN (2005). Results a. 64.29% of the respondents agreed to the use of detergent in soaking before washing, 25.14% do not use detergent in washing their carrots before selling to consumers while 10.57% were indifferent. Anionic Surfactant Residue in Exposed Carrots There was a concentration and time dependent increase (P < 0.001) in the percentage anionic surfactant in the exposed carrots Figure 1: Anionic Surfactant present as Residue in Carrot Washed with Detergent. b. Percentage Cationic Surfactant Residue in Exposed Carrot The percentage cationic surfactants residue increased with concentration and length of exposure. Figure 2: Percentage Cationic Surfactant Residue in Exposed Carrot. Figure 29: Quantity of Detergent Residue in Exposed Carrot. Conclusion The presence of residual amount of detergent in the exposed carrot raises a public health concern as this food item is daily consumed by unsuspecting public.

Environmental risk assessment in time and space - new approaches to deal with ecological complexity (P)

MO218 Uncertainty concepts and misconceptions for landscape scale risk assessment P. Thorbek, Syngenta / Environmental Safety; M. Hamer, Syngenta / Environmental Safety; K.Z. Travis, Syngenta / Product Safety; A. Raybould, Syngenta
In Europe there appears to be two opposing views on the future of ecological risk assessment of pesticides. One aims to improve ecological realism and move towards landscape scale risk assessments whereas the other aims to simplify and focus on lower tier exposure and effect assessment. To some extent this dichotomy is based on differences in uncertainty perception. Thus, one camp focuses on what we here term “in-study uncertainty” and therefore holds that higher tiers are more uncertain than lower tiers. The other camp focuses on what we term “extrapolation uncertainty” and therefore holds that higher tiers are closer to real uses in real landscapes than the lower exposure tiers. A purely statistical view of uncertainty often assumes different sources of uncertainty are independent and when this is combined with a focus on high percentiles, uncertainty measures quickly multiply up and inflates the perceived uncertainty. However, biological systems are controlled by a range of feedback and regulating mechanisms aimed at maintaining homeostasis and ecosystems normally have at least some redundancy; therefore uncertainties are typically not multiplicative. Further complicating the different views of uncertainty, is the natural variability in real landscapes. One view holds that the effects of pesticides should be isolated from the natural variability to describe the “true” effect and, since this is difficult, holds that landscape scale risk assessments increases uncertainty. The other view holds that the effect of pesticides should be related to the natural variability and hence landscape scale risk assessments reduces the other alternative’s view of uncertainty. Here we compare the two different components of uncertainty, what role they play in landscape scale risk assessment and we propose a way forwards for making uncertainty analysis more useful for decision making.

MO219 Concept for a regional geospatial landscape analyses to predict site specific vegetation covers A. Toschki, Research Institute gaia; G. Lennartz, Research Institute gaia / gaia - Research Institute for Ecosystem Analysis and Assessment; T. Schad, Bayer Ag / Environmental Modelling; T. Preus, Bayer Ag / Environmental Safety
The spatial pattern of plant communities in the agricultural landscape is depending on land management and the site specific environmental and soil conditions. In vegetation science the composition of plant species and their abundances in dependence on these factors are extensively described. This project follows the hypothesis that if the decisional environmental and soil parameters (soil type, soil texture, moisture etc.) are known, this is possible to predict the vegetation cover and subsequently the composition of plant species on this site. As a starting point the main grassland types of North Rhine-Westphalia and Mecklenburg-Western Pomerania (Germany) were considered and data for vegetation communities, plant species and their frequency and abundance were imported in a PostGis database. Additionally geospatial data (shapes of grasslands, soil types etc.) were imported in this spatial database. As a second step a matrix of combinations of soil and environmental parameters was built and calibrated in ‘if-then’ steps with the main preferences of the different vegetation communities. The poster show first prediction results and discuss pro and cons of the concept as well as possible refinements in the future. The supply of data originated from these predictions could be helpful in many facets of risk assessment on a regional scale.

MO220 B-Rice: bird focal species identification in rice paddy A. Caffi, ICPS / International Centre for Pesticides and Health Risk Prevention / Public Health; F. Marchetto, ICPS / Public Health; F. Galimberti, A. Riva, ICPS / International Centre for Pesticides and Health Risk Prevention / Public Health
Pesticide registration in EU (Reg. 1107/2009 EC) requires appropriate risk assessment for non-target organisms including birds. The European Food Safety Authority developed a Guidance Document (GD) to conduct the risk assessment considering a series of exposure scenarios from a combination of crops and growth stages, selecting relevant species at the lower steps of a tiered approach. The actual GD doesn’t include scenarios for pesticide applications on rice; nowadays bird risk assessment is generally performed considering rice as the other cereals. Rice paddy is characterized by two cultivation periods: the dry one, comparable to bare soil scenario (as common cereal), for which groups of species are equivalent to those identified for the actual risk assessment and already reported in GD; the flooded one, typical of aquatic environments and wetlands, representative of a unique exposure scenario not yet considered in the employed GD. The aim of this work is to characterize areas of rice growing in Northern Italy, which are representative for humid scenarios (via GIS approach), identify and link the relevant focal species to them. A review of the grey literature will be performed in order to estimate presence, abundance, dominance and diet of species associated to North of Italy rice paddies. Indicator and generic focal species will be proposed for the lower tiers of a Specific Rice Pesticide Risk Assessment and suggested as potential model for the Southern European Zone.

MO221 A process-based population model for algae L. Azevedo, BASF SE, Agrarzentrum Limburgerhof / Global Toxicology; M. Hübbeck, BASF Corporation; P. Jansz, BASF SE Agrarzentrum Limburgerhof
EFSA’s guidance document for the risk assessment of edge aquatic organisms recommends a maximum of 8 weeks for the ecological recovery option (ERO) of aquatic organisms in a risk assessment for plant protection products (PPP). Here, we propose a process-based model for algae abundance to simulate effects and recovery of algae populations over time following exposure to PPPs. The model integrates the main processes driving algal cell growth, such as (1) toxicity of the PPP and (2) growth limitations due to dry one, comparable to bare soil scenario (as common cereal), for which groups of species are equivalent to those identified for the actual risk assessment and already reported in GD; the flooded one, typical of aquatic environments and wetlands, representative of a unique exposure scenario not yet considered in the employed GD. The aim of this work is to characterize areas of rice growing in Northern Italy, which are representative for humid scenarios (via GIS approach), identify and link the relevant focal species to them. A review of the grey literature will be performed in order to estimate presence, abundance, dominance and diet of species associated to North of Italy rice paddies. Indicator and generic focal species will be proposed for the lower tiers of a Specific Rice Pesticide Risk Assessment and suggested as potential model for the Southern European Zone.

MO222 Population dynamics of a soil arthropod simulated using an individual based population model and established fate model data D. Nickisch, T. Wittwer, INPL / Environmental Physics Research Institute for Ecosystem Analysis and Assessment; T. Schad, Bayer Ag / Environmental Safety
The prediction of concentrations of plant protection products in soil, surface and ground water using chemical fate modelling is established since decades and applied in European environmental risk assessments (ERA). Many issues, concerns
and limitations were discussed and adequately addressed by scientific communities, applicants and authorities. As a result, a variety of accepted tools are now available for exposure assessment. In contrast, population modelling approaches used in ERA for effect assessment in ecotoxicology are still at a relatively early establishment stage. In this respect, although effect modelling is recognized as a beneficial tool for adding ecological realism to ERAS, EFA has so far not published any guidance document. Population modelling is at least mentioned in several EFSA (draft) guidelines and opinions as a refinement option. However, so far, outcomes of submitted effect modelling approaches in ERAs are not often considered by authorities. Many of the available models deal with protection goals that address field populations. Therefore, effect models are often stochastic and spatially explicit. This however makes these models more complex in comparison to the established deterministic exposure models and therefore considerable effort is needed for their verification, validation and comprehensive communication. Since effect modelling for ERA aims to predict effects on populations of the model organisms which arise from environmental exposure, we find it meaningful to use, in ecological models, the relevant data from the established fate models. This can make the modelling approaches more harmonized and probably would enhance their acceptability. We illustrated the usability of data on environmental conditions which agree with the established fate models and could as well demonstrate the implications of different environmental conditions on springtail populations. For this purpose, we used an individual based population model which represents the life-cycle of springtails in a temperature dependent framework. Specifically, we calculated soil temperature series with the groundwater model Pearl. Further, we used soil moisture conditions in functions of DOC concentrations after variable pre-equilibration times from JRC databases. Those series characterized the environment in the springtail model and drove the temperature-dependent model processes.

MO223 Dynamic modelling of fluxes of weathered polychlorinated biphenyls (PCBs) in soil: column experiments vs. modelling approaches in realistic environmental conditions

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A column leaching experiment was performed to simulate realistic conditions that may be representative of different environmental scenarios and evaluate their influence on mobility and transport associated to dissolved organic carbon (DOC) and fine particles of aged polychlorinated biphenyls (PCBs) (PCB 28, 52, 101, 153, 138, 180, 209) in soil obtained from the Brescia-Caffaro contaminated site. The concentrations measured in leachates were compared to the results of simulations performed with a dynamic, air-litter-soil model (SoilPlus model) to investigate the predictive ability and the improvements needed to obtain better predictions. The variables taken into account were: 1) contact time between soil and water, 2) DOC content and quality in leaching solutions and in soil pore water, 3) fine particle-mediated transport, 4) temperature (and its influence on endogenous DOC production), 5) soil saturation conditions. These conditions were evaluated with regard to their potential influence on the mobility and transport of PCBs under equilibrium conditions (25 °C vs. 15°C) and in saturated vs. pseudo field capacity conditions. Results indicated that equilibrium time determined differences in measured PCB concentrations up to a factor of 8, probably due to the lack of equilibration with the endogenous DOC in DOC equilibrium fractions. The addition of exogenous DOC incremented mobility (up to a factor of 4) especially for brief contact time (non-equilibrium conditions) and within the Log Kow range 6-7.5. Samples leached at room temperature showed concentrations up to a factor of 9 higher in comparison to samples collected at lower temperature probably because of the different amount of endogenous DOC produced. Samples kept in pseudo field capacity conditions for seven days and then flushed resulted in about double the concentrations of the samples flushed in saturated conditions with a brief contact time, showing that drying-wetting cycles may determine concentration peaks. These trends were not caught by the model predictions as well as the relevance of the transport associated to fine particles, pushing for incorporation of this dynamic in models.

MO224 Assessing the trait-based ecological vulnerability of aquatic invertebrates for phenol

J. Park, Gwangju Institute of Science and Technology / School of Earth Science and Environmental Engineering; R. Kim, School of Earth Science and Technology; J. Kim, S. Kim, Gwangju Institute of Science and Technology / School of Earth Science and Environmental Engineering

Recently, there has been considerable concern about the chemical accidents as usage and manufacture of phenol have increased. Phenol is harmful to living organisms and its exposure can cause ecological and economical damages. Therefore, it is necessary to prepare for possible chemical accident of phenol. The Korean government designated phenol as the accident preparedness substance and required to assess and manage the ecological risk of phenol. This study aimed to find the ecological risk at the scenario that phenol exposed to freshwater bodies in Korea. In particular, the vulnerability of aquatic invertebrates was explored to consider not only individual organisms also their populations. Vulnerability integrated the exposure, sensitivity, and recovery of the ecosystem by considering various traits (e.g., body length, food preference, toxicological sensitivity, recovery strategy, etc.). The traits were reviewed by published data or open sources, and respective scores were assigned by using multi-criteria analysis which transformed the diverse numerical entries. The toxicological sensitivity was derived by indirect prediction based on traits because base data was not possible. The results figured out the vulnerable invertebrates for phenol in Korean freshwater. In addition, the vulnerable species showed that the consideration of only sensitive species would not be great ecological risk assessment and management. This work was supported by Korea Environmental Industry & Technology Institute (KETI) through "The Chemical Accident Prevention Technology Development Project", funded by Korea Ministry of Environment (MOE) (No. 201600197001).

MO225 Assessing and managing food-web effects of Plant Protection Products

K. Swarowsky, German Federal Environment Agency (UBA) / Department IV plant protection products; H. Hötker, Nature And Biodiversity Conservation Union (NABU) Germany / Michael-Otto-Institute; R. Oppermann, Institute for Agro-ecology and Biodiversity (IFAB); C. Bruehl, University of Koblenz-Landau / Institute for Environmental Sciences; S. Matecki, German Environment Agency UBA; J. Wogram, German Environment Agency UBA / Department IV plant protection products

Assessing impacts on biodiversity needs to integrate indirect effects ( trophic chain interactions, also referred to as food-web effects or effects on biodiversity). Plant protection law requires protecting biodiversity and data requirements for Plant Protection Product (PPP) active substances (Regulation EC 283/2013) also acknowledge this aspect by mentioning indirect effects to be considered in the assessment of the impact of applied PPPs. The regulatory framework of PPPs has been well documented and recent scientific opinions of the European Food Safety Authority (EFSA) confirm the need for their inclusion in the environmental risk assessment of plant protection products. As a first step towards this direction, the currently renewed approval of glyphosate includes an obligation to the EU Member States to assess and manage the risk to diversity and abundance of non-target terrestrial arthropods and vertebrates via trophic interactions in the course of authorization procedures of glyphosate products. Therefore, we consider it necessary to develop an extension of the risk assessment to evaluate the indirect effects of specific PPPs in addition to the standard risk assessment and provide suggestions to risk managers on how to mitigate them. Due to the large variation in trophic compositions and spatial and temporal implications, we do not consider it possible to achieve a representative and realistic estimate of indirect effects by means of mechanistic models. Instead, we suggest a simple empirical model to complement current risk assessment. However, implementing a risk assessment scheme for an additional subject of protection would not be feasible without offering solutions on how to manage the assessed risk. Otherwise, an adverse outcome of this assessment would inevitably lead to non-authorisations. To solve this conflict, we put forward an approach to manage risk by means of compensating food web effects. In practice, compensation is established by ecological compensation areas such as flowering margins, set-asides and beetle banks infielf. To make most out of existing types of suitable measures and to enable a maximum of freedom of choice to farmers, we provide a points rationing scheme to categorise the individual measures with regard to their value for supporting in-field biodiversity (and thus to compensate for indirect effects of PPPs).

MO226 Compensating for ecological risks of pesticides

S. Matecki, K. Swarowsky, German Environment Agency UBA; J. Wogram, German Environment Agency UBA / Department IV plant protection products

Current environmental risk assessment (ERA) of pesticides overlooks a considerable part of existing risks and consequently fails to protect the environment from pesticide effects in toto. Examples of such blind spots are risks to field-dwelling species including wild pollinators, amphibians and farmland birds as well as indirect food web effects. To address these, we present a method which was scientifically well described and highly relevant for the achievement of the legally defined environmental protection goals, eliminating these blind spots in the risk regulation has failed so far. The ‘indirect effect’-issue is an illustrative example for what we would actually regard as a crisis in environmental risks regulation of PPPs. What we recognize is that progress in ERA notably seems to be hampere for types of risk for which no effective risk assessment procedures have been established, so that an assessment of such risks would inevitable lead to non-authorizations. To solve this conflict, we wish to put forward a radically new approach in risk management: Compensating adverse effects of pesticides where established methods of risk mitigation fail to prevent them. Once implemented into the iterative process of risk assessment, such new risk mitigation approaches would allow to manage actual risks more adequately than currently possible, thereby preventing an increase of non-authorizations. It has not escaped our notice that our proposal could also make excessive higher tier assessments dispensable, thereby helping to solve the problem of the increase of complexity in ERA.
Fish model species in human and environmental toxicology (P)

MO228

Historical control data of the optimized Zebrafish Embryo Developmental Toxicity Assay (ZEDTA)

D. van den Oetelaar, Charles River Laboratories Den Bosch / GIT; M.A. Tobor-Kaplon, Charles River Laboratories Den Bosch / GET; M. Beekhuijzen, H. Emmen, Charles River Laboratories Den Bosch / GIT; B. van de Waart, Charles River Laboratories Den Bosch / GET

The ZEDTA is a promising and innovative method with a potential to replace the screening of teratogenicity in animals (rats and rabbits) and is gaining acceptance among scientists and regulators. However, so far no harmonized and validated protocol exists for this test. Therefore, a protocol based on the OECD guideline No. 236, has been developed and optimized by Charles River Laboratories Den Bosch, the Netherlands. Multiple studies were performed using the optimized protocol, which allowed for collection of historical control data on the frequency of malformations, mortality and development of the embryos/larvae exposed to the control treatment. In our protocol, embryos in the blastula phase (2-4 hours post fertilization (hpf)) were selected and exposed to adjusted ISO medium. Twenty four embryos (one embryo per well in a 24 wells plate), were exposed at a temperature of 26°C for a period of 96h. Exposure medium was renewed after 48 hours. Development was assessed daily using the Extended General Morphology Score (GMS). Teratogenic endpoints were scored as ‘present’ or ‘absent’ after 96 hours of exposure. The Extended GMS grades the normal development of a zebrafish embryo up to 100 hpf. Assessed endpoints consisted of, but were not limited to: detachment of tail, somite formation, eye development, heartbeat and movement. The maximum score at 100 hpf was 18. Teratogenic endpoints scored during the assessment comprise of malformations of saccule/otoliths, head, heart, tail, yolk, pectoral fins and entire body. Data of twenty experiments were analysed. In total 400 embryos were exposed to control treatment (i.e. adjusted ISO medium). The average mortality rate in the control treatment was 2.5% which was considered acceptable. In only three experiments a maximum mortality of 10% was reached, which was still considered acceptable. Sixty percent of surviving larvae scored the maximum of 18 points for development, whereas 32% scored 17 points at the end of exposure (100 hpf). The most frequently observed findings were malformations of yolk (3.3%) tail (3.1%), heart (2.3%) and head (1.3%). These findings were observed in 6.4% of surviving larvae only. Analysis of the historical control data shows that the used optimized protocol produces an optimal development rate of exposed embryos and larvae, with minimal mortality and a minimal background malformation rate. This indicates a low level of confounding factors and high reliability of results produced with our protocol.

MO229

Optimization of the Zebrafish Embryo Developmental Toxicity Assay (ZEDTA)

D. van den Oetelaar, Charles River Laboratories Den Bosch / GIT; M.A. Tobor-Kaplon, Charles River Laboratories Den Bosch / GET; M. Beekhuijzen, H. Emmen, Charles River Laboratories Den Bosch / GIT; B. van de Waart, Charles River Laboratories Den Bosch / GET

The ZEDTA is a promising and innovative method with a potential to replace the screening of teratogenicity in animals (rats and rabbits) and is gaining acceptance among scientists and regulators. However, so far, no harmonized and validated protocol exists for the ZEDTA. The aim of this research was to optimize the protocol, i.e. examine which combination of exposure parameters is optimal for embryonic and larval development and is at the same time most cost-effective. An optimal embryo length should support normal growth and development with minimal mortality and/or malformations. The OECD guideline No. 236 was used as base. In our protocol embryos in the blastula phase (2-4 hours post fertilization (hpf)) are exposed to adjusted ISO medium. The following factors and their combinations were investigated: temperature (26 vs. 28°C), exposure vessels (24 vs. 96 well plates), renewal periods (static (24 or 48 h renewal)), and use of solvent (0.05% v/v DMSO vs. adjusted ISO medium). Development was scored daily, using the Extended General Morphology Score (GMS). This system grades the normal development of a zebrafish embryo up to 100 hpf. Assessed endpoints consist of, but are not limited to: detachment of tail, somite formation, eye development, heartbeat and movement. The maximum score at 100 hpf is 18. Teratogenic endpoints such as malformations of saccule/otoliths, head, heart, tail, yolk, pectoral fins and entire body were scored as ‘present’ or ‘absent’ after 96 hours of exposure. Mean developmental and teratogenic scores were calculated and used to select the most optimal condition for each factor. Our experiments showed that exposure in 24-well plates at a temperature of 26°C in combination with renewal of exposure medium after 48 hours of exposure produced the most optimal results with the lowest incidence of malformations. Daily renewal of medium provided similar results, but this was less cost-effective. Use of 0.5% v/v DMSO did induce more malformations or mortality than exposure to adjusted ISO medium.

MO230

Reliability of ecotoxicological studies in fish

H. Winnenmann, Bavarian Environment Agency; H. Ferling, Bavarian Environmental Agency; G. Dembek, W. Schmidt, W. Koerner, Bavarian Environment Agency; J. Schwaiger, Bavarian Environment Agency / Aquatic Toxicology and Pathology

For the evaluation in ecotoxicology valid bioassays are essential for deriving Environmental Quality Standard (EQS). The generally established biotests using the three trophic levels - algae, invertebrate and fish - according to OECD Guidelines provide in particular the baseline data for the derivation of the EQS. To obtain the most accurate EQS by use of a low assessment factor of 10 data from all three trophic levels including long-term results are required. Depending on the test substance growth inhibition of algae, immobilization of daphnia as well as deformation and death of fish embryos are not necessarily the most sensitive organisms and endpoints. Fish are in many cases the most suitable test organisms to demonstrate effects of e.g. pharmaceuticals with a specific mode of action in vertebrates. Therefore, prolonged toxicity tests with fish are of great importance.

However, the study design has to be adapted to specific endpoints according to the pharmacodynamics of the tested drugs. Thus, in planning and implementing this type of study special care must be taken to ensure that the generated data can be used for derivation of EQS. Even though several reporting and evaluation criteria for ecotoxicological studies have been published (e.g. Klimisch, Cred) up to date still numerous studies are available which do not fulfill these criteria and thus have to be excluded in the assessment process. Frequent failure sources include e.g. not reporting of significant p-values, thus increasing the sensitivity of results and considering so far neglected aspects such as possible background contamination of commercially available fish feed frequently used in fish studies.

MO232

Assessment of the relationship between heavy metal bioaccumulation and biomarker responses in Japanese dace inhabit in heavy metal contaminated river

H. Takeuchi, Toyo University / Research Center for Life and Environmental Sciences; Y. Iwasaki, National Institute of Advanced Industrial Science and Technology AIST Japan / Research Institute of Science for Safety and Sustainability; D. Kitamura, Toyo University / Graduate School of Life Sciences; Y. Kato, Toyo University / Faculty of Life Sciences; Y. Shimizu, Toyo University / Graduate School of Life Sciences; H. Tatsuta, University of the Ryukyus / Faculty of Agriculture; S. Kashiwada, Toyo University / Graduate School of Life Sciences The Watarase River, running in the northern Kanto region of Japan, had been severely polluted by heavy metals due to Ashio mining activities from late 1800s to early 1900s e.g. 20 mg Cu/L in river water in 1897. Through the heavy metal concentrations remarkably decreased since 1960s, the concentrations are still higher than those in unpolluted river, the Omoi River. In previous study, our group investigated the heavy metal accumulation status (Cu, Zn, As, Pb, Cd and Fe) in organs of Japanese dace Tribolodon hakonensis captured from mid reach sites in the Watarase and Omoi River, and also analyzed those river water and sediment concentrations. Water and sediment concentrations remarkably decreased since 1960s, the concentrations are still generally higher than those in the Omoi River, interestingly, whereas Zn, As, Pb and Cd concentration in liver of Japanese dace in the Watarase River were lower than those in the Omoi River. Additionally, although there were no significant genetic differences between both riverine dace in microsatellite analysis, bile metallothionein (cysteine-rich protein involved in metal detoxification) concentration of dace in the Watarase River was significantly higher than that in the Omoi River. It suggests that the dace inhabit in the Watarase River may have been adapted to metal contamination by biological responses not depending on genetic characteristics. In the present study, for understanding biological response mechanism of dace to the metal contamination, we analyzed multiple biomarkers (such as erythrocyte 6-aminolevulinic acid dehydratase activity, blood protoporphyrin and hemoglobin concentration, and bile metallothionein concentration) in dace captured in the Watarase and Omoi River, as well as metal accumulation status of those fish. In this presentation, we will show detailed results, and discuss about the relationship between heavy metal bioaccumulation and biomarker responses in riverine fish inhabit in metal contaminated river.
the objective of this study was to determine the toxic and genotoxic effect of the metals Cd, Cr, Cu and Pb which are present in the freshwater systems of the valley of Mexico, on juveniles of D. rerio, to evaluate the use of these fish as biosensors in environmental monitoring studies. Static bioassays were carried out with a duration of 48 hours, with each metal. Five toxic concentrations were used in duplicate, plus a non-toxic control. The LC₅₀ was determined and with the surviving organisms the evaluation of genetic damage was carried out, by means of the evaluation of frequency of micronuclei in blood cells (1000 cells). The results obtained showed that the toxicity of metals and their mixture, based on the LC₅₀ calculated was: Cu> Pb> Mix > Cr > Cd. The Kruscal-Wallis test indicated that there are significant differences between the degree of genetic damage in exposed organisms to different metals and controls (0.05%). The metal with the highest genotoxic effect was lead (0.05%), followed by cadmium (0.05%). Copper showed the lowest genotoxicity (0.37%). The metal mixture had a microunucleus frequency of 1.23 %. The juveniles of D. rerio had deleterious effects in concentrations of metals lower than the LMP (Maximum Permissible Limits) that marks the NOM 001 Semarnat for water discharges to natural systems, so it is possible that they can be used as biosensors in the studies of environmental monitoring. 

MO233 Endocrine disruption effects of bisphenol S and bisphenol SIP in adult zebrafish (Danio rerio) K. Li, J. Lee, Yongin University As alternative compounds of bisphenol A (BPA), bisphenol S (BPS) and 4-hydroxy bisphenol A (4OHBPA), bisphenol Sphenyphosphoryl sulfone (BPSIP) are widely used in thermal paper products. These compounds have been detected in human urine samples; however limited information is available on their endocrine disrupting effects. Adult zebrafish pairs (Danio rerio) were exposed to environmentally relevant concentrations (0, 0.5, 5, and 50 μg/L) of BPS and BPSIP for 21 days, and the adverse effects on egg production, levels of sex steroid hormones, and transcription of genes related to hormone regulation of the atria and heart (HGG axis) were investigated. The estrogenic (increase in 17β-estradiol/testosterone [E2/T] ratio) and androgenic (decrease in T) effects were commonly observed in zebrafish exposed to BPS and BPSIP, and males were more sensitive to the adverse effects than females. Although the effective concentration for endocrine disruption was greater than that of BPS, the actions of BPSIP on the steroidalogenic pathway were similar to the effects of BPS exposure. The commonalities and differences in the toxicity of BPS and BPSIP can be explained by their chemical structure: the phenolic hydroxy group is the key structural component responsible for the estrogenic and anti-androgenic activities of bisphenol analogues. The results of the present study showed that exposure to low level BPS and BPSIP could affect regulatory systems of HPG axis in zebrafish at environmentally relevant concentrations.

MO234 Oxidative Stress Induced by PAH Metabolism: Comparing Three Exposure Routes in Red Drum, Florida Pompano, and Southern Flounder to DWH surrogate oil. D. Wetzel, Mote Marine Laboratory / ELF, R. Medvecky, C. Miller, K. Main, T.A. Sherwood, Mote Marine Laboratory The magnitude of the oil and dispersant released during the Deepwater Horizon blowout caused significant immediate, and often lethal, damage to exposed organisms. However, the sub-lethal impacts of the chronic spill on offshore and nearshore biota are still not fully characterized. To help understand one of the most significant responses in important Gulf fish species, four different exposure-route experiments, DWH surrogate oil contaminated feed, sediments, and seawater, were designed and carried out to examine biological responses of aquaculture reared red drum, Florida pompano, and southern flounder. Environmental pollutants, like polycyclic aromatic hydrocarbons (PAHs) found in crude oil, have the potential to regulate the antioxidant system of marine organisms. Oxidative stress occurs when there is an imbalance between the production of reactive oxygen species (ROS) and the organism’s ability to detoxify reactive intermediates, such as those generated by metabolism of PAHs by cytochrome P450 (CYP1). Depending on the severity of oxidative stress, this imbalance can lead to DNA damage in a variety of ways, such as oxidized bases, apurinic/apyrimidinic sites (AP sites), single or double strand breaks and DNA adducts. Exposure to PAHs can lead to increase DNA damage, such as those created by AP sites (purine loss) and the formation of DNA adducts, in which PAH metabolites intercalate into the DNA. Total PAH concentrations were analyzed in exposure matrices, as well as fish livers and whole bodies to determine specific dosages. Multiple assessments have been carried out to examine oxidative stress, total antioxidant power analysis, 2-Thiobarbituric Acid Reactive Substances analysis, GSH/GSSG ratio determination, AP site quantitation, and 8-OHdG quantitation. Evidence of oxidative stress will be discussed comparing multiple pathways of exposure, and resulting impacts in terms of biological and ecological implications.

MO235 Impact of PAH/oxo-PAH mixtures on heart development in zebrafish V. Cunha, K. Dreij, Karolinska Institutet Polycyclic aromatic hydrocarbons (PAHs) are ubiquitous environmental pollutants that have been widely studied. Oxygenated PAHs (oxy-PAHs) are also found in the environment and are emitted from the same primary sources as PAHs but also can be formed through secondary oxidation of PAHs. However, relatively little is known about their environmental fate and toxicity. The aim of this work was to determine the effects of binary PAH/oxo-PAH mixtures on cardiac development in zebrafish (Danio rerio) embryos (ZFEs). ZFEs [24 hpf] were exposed to a dose range of single PAH (benzo[a]pyrene, BP), oxy-PAH (the ketones 4H-cyclopenta[d]anthracene-4-one (4H-CPO), benz[a]fluorenone (BFLO) and 6H-benzo[c]pyrene-6-one (6H-BPO)) or their binary mixture for 4 days. After exposure, ZFEs were observed for effects on heart development, heart rate and blood flow. Samples were also collected for gene expression analysis. The results showed abnormal cardiac development, such as formation of string hearts where exposure ranged from 6H-BPO and BFLO in combination with BP was more potent than single exposures. The heart rate and blood flow was significantly decreased, in a dose-dependent manner, in ZFE exposed to 6H-BPO alone and in combination with BP. With the other oxy-PAHs, a decrease in the heart rate was observed, however in a non-monotonic response to the treatments. Blood flow was also decreased but only for BFLO and 4H-CPO in mixture with BP. Gene expression analysis showed significant up-regulation of genes involved in cardiac development (trh5), especially for ZFE exposed to the combination of oxy-PAHs with BP. Notably, the up-regulation of these two genes correlated with the formation of string heart. In summary, the binary mixtures were more potent then oxy-PAHs alone in inducing cardiototoxicity, except in the case of 6H-BPO which seems to be a very potent oxy-PAH. The oxy-PAHs and PAHs interact and thereby influence the other effects simultaneousizes the importance of monitoring the presence of oxy-PAHs in the environment.

MO236 Induction of developmental cardiotoxicity in rainbow trout (Oncorhynchus mykiss) following PAH mixture exposure - new insights using an integrated OMICS approach. A.N. Eriksson, C. Rigaud, University of Jyväskyla / Department of Biological and Environmental Science; A. Krasno, NOFIMA; J. Liibavainen, University of Helsinki; A. Ronkka, S. Saraiu, T. Suomi, A. Laiho, L. Elo, University of Turku and Åbo Akademi University; E. Vehniäinen, University of Jyväskyla / Department of Biological and Environmental Science Worldwide, environmental levels of polycyclic aromatic hydrocarbons (PAHs) have increased over the last century, mainly due to anthropogenic release from incomplete combustion of organic material and oil spills. These compounds are present as a complex mixture in the environment and are known to cause developmental abnormalities, cardiotoxicity, immunosuppression, tumors and alteration of reproductive performance. Many PAHs, such as phenanthrene, have been studied for over 100 years and it is currently known that different PAHs have different modes of action (MoA). PAHs like retene and pyrene are aryl-hydrocarbon receptor agonists that up-regulate CYP1a expression and thereby induce their own metabolism, while other PAHs like fluoranthene directly inhibit CYP1a activity. In 2014, we exposed newly hatched rainbow trout fry (Oncorhynchus mykiss) semi-statically to retene and fluoranthene to either alone or as a mixture treatment for 1, 3, 7 and 14 days. Body length and yolk (energy) consumption were measured and the hearts collected and pooled at the end of every exposure period. Using transcriptomics, proteomics and metabolomics, cardiotoxicity was investigated at molecular level. Our results shows that fry exposed to retene and the mixture treatments, in relation to control, became shorter and in retene’s case, had used more yolk by day 14. Fluoranthene exposure at sub-lethal concentration did not affect growth or energy consumption by day 14. The only gene that was significantly up-regulated in the surviving organisms was the MoA gene CYP1a. Using overrepresentation analysis, several pathways were affected by PAH exposure, but only limited information is available on their endocrine disrupting effects. Northern analysis revealed several pathways affected by PAH exposure, except in the case of 6H-BPO which seems to be a very potent oxy-PAH. The oxy-PAHs and PAHs interact and thereby influence the other effect simultaneousizes the importance of monitoring the presence of oxy-PAHs in the environment.
includes detoxification enzymes induction (CYP1A), hemorhaging, cardiovascular defects, pericardial and yolk sac edemas, craniofacial deformities or growth attenuation. The cardiovascular tissue is one of the most sensitive to PAHs, and all the aforementioned symptoms are caused by the activation of the aryl hydrocarbon receptor (AhR). However, the mechanisms involved downstream of the AhR activation by PAHs are still unclear. Some weak AhR agonists such as phenol can also activate this receptor (effectives, e.g. arfiphenine) via unknown AhR-independent mechanisms. In this study, we aimed to explore the mechanisms of toxicity of individual PAHs in the rainbow trout (Oncorhynchus mykiss) ELS by the use of an integrated OMICS approach, i.e. the combined use of transcriptomics, proteomics and metabolomics. The use of OMICS can lead to evidence of which pathways are altered by PAHs, and thus help choosing candidate genes as biomarkers of their mechanism of toxicity. Newly hatched rainbow trout larvae were exposed to three different PAHs (retene, pyrene or phenanthrene) at sublethal doses. The heart of each larva was sampled after 1, 3, 7 or 14 days of semi-static exposure, and RNA, proteins and metabolites were extracted. Morphometric parameters such as larval length and yolk sac area were also monitored, but were rarely affected by PAHs. Preliminary data from transcriptomics and metabolomics showed different signatures of gene expression alteration as well as different metabolite profiles between treatments, suggesting specific mechanisms of toxicity. Overall, all compounds induced more changes in cardiac gene expression during the very first days of development, with the exception of pyrene which was also very potent after 7 days. Preliminary enrichment analysis (over-representation analysis) revealed that differentially expressed genes were interchangeable in their biological functions. Exposure of the embryos by cyanethane appeared to alter collagen biosynthesis, as well as the glutamate release cycle, but only at one sampling point and with very few genes involved. Proteomic analyses are underway to further highlight the mechanisms of toxicity.

**MO238**

**Developmental Toxicity of a Non-steroidal Anti-inflammatory Drug (Acetaminophen), in African Catfish (Clarias gariepinus) embryos.** L.1 Ezemanye, University Benin / Animal and Environmental Biology; N.O. Ezekwe, University of Benin, Benin City, Nigeria / Animal and Environmental Biology; I. Tongu, University of Benin / Laboratory of Ecotoxicology and Environmental Forensics, Faculty of Life Sciences, Department of Animal and Environmental Biology,University of Benin, Nigeria; P. Adebayo, University of Benin / Animal and Environmental Biology.

The study investigated the developmental toxicity of Acetaminophen, a non-steroidal Anti-inflammatory Drug on the early life stage (0 to 96 hpf) of African Catfish (Clarias gariepinus). The 96 hrs fish embryo acute toxicity (FET) test was carried out according to the modified OECD 236 guidelines. Newly fertilized embryos were exposed to different concentrations (0, 0.5, 1, 5 and 10 µg L⁻¹) of the drug in triplicates and observations of embryo development were made at different developmental stages. Morphological, physiological and behavioural malformations were observed with its exposure. Exposure of the embryos by acetaminophen significantly altered the morphological and behavioural properties of the fish. The effects were observed to be dose and time-dependent, as more poisoning symptoms were recorded at higher dose. Exposed embryos were observed to have poorly formed somite’s, coagulated embryos, non-detached tail, altered spontaneous movement and inhibited swimming performance. Observed physiological alterations include cardiac edema, sac yolk edema, pericardial edema, tail malformation and lordosis. The result demonstrated that acetaminophen has the potential to alter the development of the early life stage of the African catfish.

**MO239**

**In vitro approach for the identification of early warning biomarkers, related to exposure to PBDEs, in human and marine systems: oxidative stress, toxicity and cell cycle modulation.** C. Espinosa, S. Manuguerra, M. Morghese, UniPa / DiSTeM; A. Cuesta, M. Irstea Lyon; R. Casadio, University of Bologna / Depar...
which is vital for energy production and is located in mitochondrial membranes. COX activities from zebrafish (Danio rerio), Corydoras (Corydoras aeneus), earthworms (Eisenia fetida), and the lesser rice weevil (Sitophilus oryzae) were placed to be inhibited by pyrogallol and its related chemicals such as gallic acid, 1,2,4-benzenetriol, pyrocatechol, caffeic acid, quinic acid, and chlorogenic acid. For the inhibition of zebrafish COX, pyrogallol was the strongest chemical among the four tested compounds, with a concentration of 20 ppm. Gallic acid and 1,2,4-benzenetriol showed potent inhibition on the COX activity with the concentration of 100 ppm. At a 10-times diluted concentration, these three compounds showed moderate inhibition on the enzyme activities. These phenomenon were applied all of the tested animals. Pyrocatechol, caffeic acid, quinic acid, and chlorogenic acid did not show any inhibitory effect on the COX activity. Taken together, benzenetriols including pyrogallol may be caused by unexpected inhibitory effects on the animal COX activity, referring fluctuation of the energy production, and the benzenetriol moiety is essential for the inhibition on the COX activity.

MO243 Exposure to environmental concentrations of Triclosan induces oxidative stress and genotoxicity on zebrafish (Danio rerio) embryos

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Triclosan (TCS, 5-chloro-2-(2,4-dichlorophenoxy) phenol) is the most common antibacterial agent used in personal care products, including soaps, body lotions, laundry detergents, toothpastes and deodorants. For its properties it is also added to several household items such as food packaging materials, toys and textiles. Since TCS is not completely removed by Waste-Water Treatment Plants (WWTPs), it is becoming a potential worldwide pollutant and it is frequently detected in surface waters, with concentrations ranging from ng/L to µg/L. There is evidence that TCS is acutely and chronically toxic to aquatic organisms and it was already demonstrated that this chemical severely affects both zebrafish adult and embryos, causing embryotoxicity, hatching delay and biomarker alterations. Thus, the European Union (EU) has disapproved in 2016 the use of TCS in biocidal products, due to its unacceptable environmental risk. Meanwhile, consumer antiseptic wash products containing TCS can no longer be marketed in the US. The aim of this study was to investigate the adverse effects of TCS on the development of zebrafish embryos. In this paper, we showed that zebrafish embryos exposed to TCS from the first 2 h post-fertilization (hpf) until 100 hpf, exhibit pericardial edema, abnormal hatching, swim bladder deformities and gastroschisis. A significant increase in the number of dead embryos was observed at the concentrations of 100 and 1,000 ng/L. The results indicated that zebrafish embryos exposed to TCS concentrations of 10, 100 and 1,000 ng/L until 100 hpf, showed oxidative stress. In particular, the analysis of oxidative stress markers in TCS-exposed embryos showed a significant decrease in the GSH level and an increase in the lipid peroxidation levels. These results indicate that TCS exposure causes oxidative stress in zebrafish embryos and that these effects may be caused by the antioxidant activity of TCS.

MO244 Comparative study of acute toxicity of a Microcystis aeruginosa bloom containing microcystin-LR on common carp (Cyprinus carpio) and Wistar rat

Z. Hadjer, R. Bordj, H. Nasri, Laboratory of Biodiversity and Ecosystem Pollution, University of El Tarf, Algeria.; N. Bouaicha, UNIVERSITE PARIS 8

Microcystins (MCs) are hepatotoxins produced by several groups of cyanobacteria in water bodies throughout the world. Their mechanism of toxicity consists of a disruption of energy production, and the benzenetriol moiety is essential for the inhibition on COX activity.

MO245 Subchronic toxicity of a Microcystis aeruginosa bloom extract containing mainly the microcystin-LR congener on the common carp Cyprinus carpio

R. Bordi, Z. Hadjer, H. Nasri, Laboratory of Biodiversity and Ecosystem Pollution, University of El Tarf, Algeria.; N. Bouaicha, UNIVERSITE PARIS

The occurrence of harmful cyanobacterial blooms in surface waters is often associated by a production of variety of cyanotoxins that represent a hazard for human and animal health. Cyanotoxins (MCs) are the most common cyanotoxins and may be expected wherever blooms of cyanobacteria occur in surface waters and the most usual MCs, more than 100 variants have been characterized. Among these cyanotoxins, microcystin-LR (MC-LR) is the most studied congener due to its high toxicity and frequent occurrence in surface waters. The purpose of the present study was to investigate the effects of 12-week gavage of a Microcystis aeruginosa bloom containing mainly the congener MC-LR (>95%), in male and female of juveniles (200 g) of common carp (Cyprinus carpio). The fishes were then randomly assigned to three groups. Group I, the control group, received daily physiological serum (500 µL), groups II and III were daily exposed by gavage (5 days per week) to lyophilized Microcystis aeruginosa bloom dispersed in physiological serum (500 µL) containing 2 and 10 µg equivalent MC-LR/kg body weight (BW) for 12 weeks, respectively. The effects on the hepatopancreas, kidneys, intestine and gills have been evaluated by histological analysis and the biomarkers of oxidative stress: lipid peroxidation, reduced glutathione (GSH) level, glutathione-s-transferase (GST) and glutathione peroxidase (GPx) activities. The histological study showed for the two doses the presence in the male and female carps: signs of hemorrhage and lymphocytic inflammatory infiltrates in the hepatopancreas, renal glomerular deformity with lymphocytic infiltrate in the kidney, epithelial cell fission to fission fusion of intestinal villi and hypochromia in some hypertrophy in some few lamellae with the gills. The exposure of cyanobacterial bloom containing the two doses of MC-LR resulted in a significant increase of lipid peroxidation and GST activity in both male and female group. However, a significant decrease in both GPx activity and the GSH level have been observed. In addition, the results of the histological study and biomarkers of oxidative stress have shown that male fish are much more sensitive to the bloom of cyanobacteria containing microcystins than females. Key words: MC-LR, Cyanic carpio, oxidative stress, histological study.

MO246 Diluted bitumen vs. conventional crude oil: effects of developmental exposure on first- and second-generation zebrafish

D. Lyons, University of Alberta; D. Philipbert, K.B. Tierney, University of Alberta / Biological Sciences

The Canadian oil sands industry produces a heavy, viscous crude oil called bitumen. Due to its viscosity, bitumen must be diluted with natural gas condensates to create diluted bitumen or ‘dilbit’ to facilitate pipeline transport. The composition of dilbit differs greatly in chemical composition compared to conventional crude oils and the impact of dilbit exposure on aquatic organisms has not been well characterized, despite its widespread transport across North America. In this study, the effects of developmental exposures on breeding success and next generation embryos were compared between dilbit and two conventional crude oils (mixed swabbed and medium sour composite). Zebrafish embryos were exposed to water accommodated fractions of these oils from 0-7 days post fertilization (dpf) and gene expression and DNA methylation were measured at 7dpf. Exposed embryos were then grown to adulthood in clean water. These fish were bred and their embryos were collected and reared in clean water (unexposed second-generation embryos). Breeding success of the first-generation developmentally exposed fish was determined by measuring the number of pairs that spawned, number of eggs spawned, fertilization rate, and survival of unexposed offspring. Gene expression and DNA methylation were also measured in 7dpf offspring. Developmental exposure in the first generation did not affect the survival of embryos and also did not affect breeding success when compared to control, but differed among exposure groups. Some target genes were differentially expressed in developmentally exposed second-generation embryos when compared to controls indicating a heritable change in basal gene expression. This change in gene expression could potentially be due to changes in DNA methylation caused by the developmental exposure in the first-generation. Understanding what changes in DNA methylation mean for fish survival will require further study. Overall, it appears that developmental exposures to bitumen and dilbit have variable effects on first- and second-generation zebrafish embryos. Though second-generation endpoints are often overlooked, they are important to consider when evaluating the overall risk of oil exposure.

MO247 Effect of skatole and its metabolites on piscine Phase I metabolism

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Cytochrome P450 (CYP) enzymes are dominant players in metabolism of xenobiotics and a wide range of endogenous compounds. Skatole is well known mammalian metabolite, however rarely considered as environmental relevant pollutant. In fact, presence of skatole in aquatic organisms may be an indication of its toxicological risk for these species due to its biological effects. To the best of our knowledge, no studies attempted to investigate the effect of skatole and its major metabolites on piscine CYPs. The aim of this study was to identify weather skatole and its metabolites, 2-aminoacetophenone, indole-3-carbinol, 3-methylindoxyl, and 3-hydroxy-3-methylindoxyl, can interact with fish CYP isoforms. Enzyme activities for CYPs in rainbow trout hepatopancreatic tissues were measured in the presence or absence of skatole and its metabolites. Following concentrations of tested inhibitors were used: 0.5; 5 and 50 µM Skatole and indole-3-carbinol showed no inhibition potency on either CYP1A or CYP2A, 2-Aminoacetophenone, 3-methylindoxyl and 3-hydroxy-3-methylindoxyl reduced CYP1A enzyme activity by approximately 25-35%, whereas CYP2A activity remained unaltered. Physiological consequences of such inhibition for fish ability to detoxify xenobiotics remain to be elucidated. Keywords: rainbow trout, cytochromes, EROD, COH Acknowledgement - The study was financially supported by the Ministry of Education, Youth and Sports of the Czech Republic, projects CENAKVA (No. CZ.1.05/2.1.00/0001.0024), CENAKVA II (No. LO1205 under the NPU I program), by the Czech Science Foundation (No. 18-15002S) and Swedish University of Agricultural Sciences.

MO248 Linkage of gene expression patterns with in vivo endpoints: gaining deeper insights
A. Zenker, University of Appl. Sc. Northwestern Switzerland / Institute of Environmental Chemistry; N. Munz, University of Applied Sciences and Arts Northwestern Switzerland

The herbicide glyphosate and the pharmaceutical isoprinor are among the most popular chemicals that have been detected in many streams at low concentrations. Nevertheless, the ecotoxicological properties, especially of isoprinor, are largely unknown. The objective of the study was to find early detection markers by comparing in vivo effects and geneexpression patterns in the fish embryo acute toxicity (FET) test and early-life stage toxicity test (EL5). Per substance 480 individuals of zebrafish were exposed to five different concentrations (isoprinor: 0.03-0.5 mg/L; glyphosate: 0.006-0.1 mg/L) and analysed for different morphological endpoints such as spontaneous movements, heart rate, hatching success and survival. Gene expression patterns of such inhibition for fish ability to detoxify xenobiotics remain to be elucidated. Keywords: rainbow trout, cytochromes, EROD, COH Acknowledgement - The study was financially supported by the Ministry of Education, Youth and Sports of the Czech Republic, projects CENAKVA (No. CZ.1.05/2.1.00/0001.0024), CENAKVA II (No. LO1205 under the NPU I program), by the Czech Science Foundation (No. 18-15002S) and Swedish University of Agricultural Sciences.

MO249 New insights on cross-species differences in the modulation of human and zebrafish nuclear receptors by single chemicals and environmental mixtures
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In the context of contamination of aquatic ecosystems by endocrine disrupting chemicals (EDCs), this work aims to provide new insights on cross-species differences, and the modulation of nuclear receptors (NRs) and their biological responses, by xenobiotics. The use of zebrafish as model organism appeared as an efficient tool to investigate on xenobiotics effects. Our results revealed that NRs are, for some of them, differentially modulated by individual chemicals and environmental mixtures. Additionally, interaction of EDCs towards NRs cannot always be extrapolated between species because highlighting the need for further document NRs modulation between human and fish and associated responses, to improve human health and environmental risk assessment of EDCs.

MO250 Combining acute toxicity, toxicokinetics and metabolomics approaches to assess the effects of triclosan in zebrafish embryos
D. E. Damalag, National and Kapodistrian University of Athens / Chemistry; M. Agalou, Biomedical Research Foundation Academy of Athens / Developmental Biology; D. Beis, Biomedical Research Foundation Academy of Athens / Developmental Biology; M. Lamoree, VU University, Department Environment & Health / Department Environment & Health; P. Leonards, VU University, Institute for Environmental Studies / Department of Environment and Health; N.S. Thomaidis, National and Kapodistrian University of Athens / Department of Chemistry

Triclosan (TCS) constitutes a common household product ingredient, given its antimicrobial activity, and has been widely used over the past decades. It enters the sewer system and can be transported to wastewater treatment plants (WWTP), sewaters and rivers, resulting in the contamination of the aquatic ecosystem. Consequently, it is urgently needed to evaluate the potential toxic effects to aquatic organisms. The triclosan has emerged as a powerful model organism to study various aspects of developmental and cell biology, while it provides an alternative model for toxicological studies. The objectives of this study were to assess to what extent TCS induce toxicity in zebrafish embryos. In addition, we evaluated the uptake and biotransformation of TCS by zebrafish and examined whether biotransformation data could be used complementary to the concentration of the parent TCS to interpret the induced toxicity. The final goal was to establish a wide-scope targeted metabolomics screening workflow to investigate the induced toxicity in a biochemical perspective and associate the observed toxicity/phenotype with changes in molecular level. Overall, the aim was to highlight a high throughput test that could be incorporated in different screening approaches, for a comprehensive toxicity assessment of environmental stressors in aquatic organisms. The zebrafish embryo toxicity assay was used to calculate the LC50 value of TCS as well as to perform the morphological phenotyping. In addition, a liver specific fluorescent transgenic line (Tg:LFABP:GFP) was used, to evaluate TCS liver toxicity potential. Concerning the toxicokinetics and the metabolomics experiment, 96 hpf zebrafish embryos were used. Samples were collected at 5 different time intervals, from 30 s up to 24 hours post exposure (hpe). Detection and identification of tentative TCS-bio-TPs was performed through in-house developed suspect and non-target screening workflows. Bio-TPs arising from both oxidative and conjugative metabolic reactions were identified. Regarding the metabolomics part of the study, a database of over 600 endogenous metabolites (carboxylic acids, amines, nucleotides etc.) was established, covering a broad range of primary metabolism. For the toxicokinetics part, a compartment model was used to represent the toxicokinetics of TCS in zebrafish embryos. The results show that the biotransformation of TCS by zebrafish embryos is efficient in producing TCS metabolites. Moreover, the toxicokinetics and metabolomics approach showed that TCS possesses high potential as an environmental threat to aquatic organisms.
extract was then centrifuged, filtered and reconstituted in methanol. Separations were performed on C18 (2.1 mm × 50 mm, 3.5 μm particle size) using methanol (0.1% formic acid) and water as the mobile phase. Negative ion electrospray ionization and specific multiple reaction monitoring ion transitions were used to detect F2-isopSs in mucus. Mass labelled internal standards were used to monitor recovery of native compounds during sample work-up and also to quantify native F2-isopSs. Natural abundance Class III and VI F5-isopSs were measurable in Crappie (Pomoxis). This work demonstrates that mucus has the potential to be used as a non-invasive, non-lethal matrix for F2-IsopS analysis in fish.

MO252

Validation of in ovo embryo microinjection as a surrogate maternal transfer of selenomethionine in the fathead minnow (Pimephales promelas)

T. Lane, University of Saskatchewan; D. Green, K. Raes, University of Saskatchewan - Toxicology Centre / Toxicology; K. Bhum, University of Saskatchewan / School of Environment and Sustainability; D.M. Janz, K. Liber, L.E. Doig, University of Saskatchewan / Toxicology Centre; M. Hecker, University of Saskatchewan / School of the Environment & Sustainability and Toxicology Centre

Selenium (Se) is a naturally occurring trace element that is recognized as a constituent of concern in Canadian aquatic ecosystems because of its high potential to bioaccumulate and persist even after its source has been removed. All animals are susceptible to the effects of Se due to the narrow range between dietary essentiality and toxicity; however, oviparous vertebrates, especially fish, are at particular risk for the embryonic and maternal transfer of S. Se. This study was conducted to develop and validate an embryo injection approach to model maternal transfer of selenomethionine (SeM), the primary form of Se in the diet. This model could then be applied to any egg-laying species of interest and could provide insight regarding differences in species sensitivity to Se toxicity during early life stage development.

Initially, the maternal transfer of dietary SeM and its effect on the F1 generation was characterized in a short-term fish species in freshwater systems, the fathead minnow (Pimephales promelas). 20 breeding groups (3 females:2 males) were fed a SeM-spiked diet of either 0, 3, 9, or 27 mg Se/kg bloodworms dry weight (dw) and bred for 28 days. Embryo Se concentrations increased immediately upon onset of exposure and Se concentrations reached approximately a 1:1 ratio in food:embryo after 28 days on the diet. There was a significant difference in mean embryo Se concentrations from the control (1.18 mg/kg embryo dw) in the medium (8.75 mg/kg embryo dw) and high (29.58 mg/kg embryo dw) treatment groups (p=0.057); however, a more robust analysis is on-going. Average Se embryo concentrations from this study will serve as the basis for subsequent embryo injection studies in fathead minnow. Developmental endpoints from both studies (e.g. mortality, frequency of deformities, types of deformities, severity of deformities) will be compared to determine if the embryo injection model is an appropriate tool for studying the maternal transfer of Se. This approach was used to establish the ontology of SeM uptake and may be responsible for xenobiotic metabolism.

MO253

Preliminary characterization of the rainbow trout intestine using omics based approaches

L.M. Langan, Plymouth University / Biological and Marine Sciences; S. Owen, AstraZeneca / Safety Health Environment; A.N. Jha, Plymouth University / Biological Sciences

Intestinal function is central to the physiology, health and disease of numerous organisms. However, little is known about its gene or protein profile in trout, a widely studied and environmentally relevant model laboratory organism (Onchorhynchus mykiss). In this study, two omics based tools focused on characterizing RNA and protein expression were used to establish the ontology of each intestinal region viz. the pyloric, anterior, mid and posterior intestine. RNA Seq was ca.

MO254

Persistent organic pollutants alter the expression patterns of epigenetic factors in the Zebrafish Liver (ZF-L) Cell line

M. Blanc, Orebro University / MTM Research Centre; N. Scherbak, Orebro University / School of Science and Technology, Life Science Centre; S. Keiter, Orebro University / MTM Research Centre

Several studies demonstrated that exposure to persistent organic pollutants can induce epigenetic modifications in human and other vertebrate tissues. It is of great interest since epigenetic changes are reported in animals with increasing incidence such as cancer. Besides, epigenetic disruption was suggested to be one mechanism responsible for multigenerational effects of chemical exposure. Epigenetic pathways in zebrafish are similar to mammalians; therefore, it was proposed as an alternative model for epigenetic research. The focus of the present study was set on the investigation of epigenetic effects in the Zebrafish Liver (ZF-L) cell line after 48 h of exposure to selected compounds. The cells were exposed to the LC10 values of pesticides (methoxychlor (MXC), permethrin (PER)), plastic additivates (bisphenol A (BPA) and S (BPS)), perfluorinated compounds (perfluorooctane sulfonic acid (PFOS), perfluorobutane sulfonic acid (PFBS)), a whitening agent, 7-dihydroxy-4-methylcoumarin (DEM); and to the diet. The cell viability value was measured using an MTT assay. Western blot analysis of expression of genes encoding enzymes and factors involved in DNA methylation and histone modifications was measured using RT-qPCR. The DNA methyltransferases were selected to target DNA methylation (dnmt1, dnmt3aa, dnmt3ab, dnmt3b). They were analyzed together with 2 histone deacetylases (hdac1, hdac3), one demethylase (bsd1l), and one chromatin remodeling factor (spth6). At the selected concentrations, all compounds, except PFBS, induced significant changes in gene expression, as measured with microarray analysis. This work demonstrates that the embryo injection approach can be applied to any egg species to develop and validate an embryo injection approach to model maternal transfer of selenomethionine (SeM), the primary form of Se in the diet. This model could then be applied to any egg-laying species of interest and could provide insight regarding differences in species sensitivity to Se toxicity during early life stage development.

MO256

Cross-species applicability of the adverse outcome pathway “deiodination inhibition leading to impaired swim bladder inflation in zebrafish”

E. Weekers, L. Vercammen, M. Blanc, K. Raes, University of Antwerp / Zebrafishlab Dept Veterinary Sciences SPHERE; H. Witters, VITO / Applied Bio & Molecular Systems; R. Blust, University of Antwerp / Department of Biology (SPHERE Research Group); G.T. Ankley, D.L. Villeneuve, U.S. EPA / National Health and Environmental Effects Research Laboratory; D. Knapen, University of Antwerp / Zebraslab Dept Veterinary Sciences

The adverse outcome pathway (AOP) framework can be used to help support the development of alternative testing strategies aimed at predicting adverse outcomes caused by triggering specific toxicity pathways. Previously, we developed an AOP describing how inhibition of deiodinase (DIO) enzyme activity leads to impaired swim bladder inflation in fish. Next we assessed the feasibility of selecting alternative in chemico assays targeting specific key events along the AOP and evaluating the potential of the well-known BPAs and PFOS. Overall, the present results showed that ZF-L cells were responsive to epigenetic disruption. They brought further evidence on the potential of chemicals to interfere with both DNA methylation and chromatin accessibility. However, further studies are required to investigate to what extent the observed changes are reflected in DNA methylation and chromatin accessibility themselves, together with their correlation in in vivo models.

MO257

Epigenetic disruption in rainbow trout cytochrome P450s under dietary conditions

M. Blanc, J. Halvorsen, L. Mortensen, R. Blust, University of Antwerp / Department of Biology (SPHERE Research Group); G.T. Ankley, D.L. Villeneuve, U.S. EPA / National Health and Environmental Effects Research Laboratory; D. Knapen, University of Antwerp / Zebraslab Dept Veterinary Sciences

Epigenetic disruption was suggested to be one mechanism responsible for multigeneration effects of chemical exposure. Epigenetic pathways in zebrafish are similar to mammalians; therefore, it was proposed as an alternative model for epigenetic research. The focus of the present study was set on the investigation of epigenetic effects in the Zebrafish Liver (ZF-L) cell line after 48 h of exposure to selected compounds. The cells were exposed to the LC10 values of pesticides (methoxychlor (MXC), permethrin (PER)), plastic additivates (bisphenol A (BPA) and S (BPS)), perfluorinated compounds (perfluorooctane sulfonic acid (PFOS), perfluorobutane sulfonic acid (PFBS)), a whitening agent, 7-dihydroxy-4-methylcoumarin (DEM); and to the diet. There was a significant difference in mean embryo Se concentrations from the control (1.18 mg/kg embryo dw) in the medium (8.75 mg/kg embryo dw) and high (29.58 mg/kg embryo dw) treatment groups (p=0.057); however, a more robust analysis is on-going. Average Se embryo concentrations from this study will serve as the basis for subsequent embryo injection studies in fathead minnow. Developmental endpoints from both studies (e.g. mortality, frequency of deformities, types of deformities, severity of deformities) will be compared to determine if the embryo injection model is an appropriate tool for studying the maternal transfer of Se. This approach was used to establish the ontology of SeM uptake and may be responsible for xenobiotic metabolism.

MO254

Persistent organic pollutants alter the expression patterns of epigenetic factors in the Zebrafish Liver (ZF-L) Cell line

M. Blanc, Orebro University / MTM Research Centre; N. Scherbak, Orebro University / School of Science and Technology, Life Science Centre; S. Keiter, Orebro University / MTM Research Centre

Several studies demonstrated that exposure to persistent organic pollutants can induce epigenetic modifications in human and other vertebrate tissues. It is of great interest since epigenetic changes are reported in animals with increasing incidence such as cancer. Besides, epigenetic disruption was suggested to be one mechanism responsible for multigenerational effects of chemical exposure. Epigenetic pathways in zebrafish are similar to mammalians; therefore, it was proposed as an alternative model for epigenetic research. The focus of the present study was set on the investigation of epigenetic effects in the Zebrafish Liver (ZF-L) cell line after 48 h of exposure to selected compounds. The cells were exposed to the LC10 values of pesticides (methoxychlor (MXC), permethrin (PER)), plastic additivates (bisphenol A (BPA) and S (BPS)), perfluorinated compounds (perfluorooctane sulfonic acid (PFOS), perfluorobutane sulfonic acid (PFBS)), a whitening agent, 7-dihydroxy-4-methylcoumarin (DEM); and to the diet. There was a significant difference in mean embryo Se concentrations from the control (1.18 mg/kg embryo dw) in the medium (8.75 mg/kg embryo dw) and high (29.58 mg/kg embryo dw) treatment groups (p=0.057); however, a more robust analysis is on-going. Average Se embryo concentrations from this study will serve as the basis for subsequent embryo injection studies in fathead minnow. Developmental endpoints from both studies (e.g. mortality, frequency of deformities, types of deformities, severity of deformities) will be compared to determine if the embryo injection model is an appropriate tool for studying the maternal transfer of Se. This approach was used to establish the ontology of SeM uptake and may be responsible for xenobiotic metabolism.
Zebrafish responses to the fourth-generation progestin drospirenone exposures

C. Quintaniero, Department of Biology & CESAM - University of Aveiro; A.M. Soares, University of Aveiro / department of Biology & CESAM; M. Monteiro, Aveiro University / Biology

Synthetic progestins (PGs) represent an important class of active ingredients of hormonal contraceptives and have been reported to exert adverse effects on fish and endocrine disruption and are in line with other studies. The present work is focused on the development in early life stages of zebrafish (Danio rerio) in response to drospirenone (DRP) exposure. DRP is a progestin that has led to increased production of trophic hormones by the hypothalamus. The hypothalamic-pituitary-thyroid (HPT) axis is known to play a crucial role in the development of testes and ovaries. Therefore, knowledge of endogenous transcription profiles of thyroid-related genes in developing teleosts remains fragmented. We selected two model teleost species, the fathead minnow (Pimephales promelas) and the zebrafish (Danio rerio) to describe the gene transcription ontology of the HPT-axis. Control embryos were sampled at several time points between fertilization and hatching, and larvae were sampled approximately every other day until 33 days post-fertilization. Total RNA was extracted from pooled, whole fish, and thyroid-related mRNA expression was evaluated using quantitative polymerase chain reaction (qPCR) experiments. The study aims to define the role of certain genes and pathways during development. Furthermore, it can function as a background reference dataset for designing and interpreting targeted transcriptional expression studies both for fundamental research and for applications, such as ecotoxicology.

MO258 Fish caging experiment as a tool for detection of in situ effects of untreated wastewaters: General stress and endocrine disruption

Department of Biology and Ecology, Laboratory of Ecotoxicology (LECOTOX); S. Kisašević, Faculty of Sciences University of Novi Sad / Biology and Ecology; V. Knezevic, Faculty of Sciences / Department of Biology and Ecology Laboratory of Ecotoxicology LECOTOX; S. Titašević, Faculty of Sciences University of Novi Sad / Biology and Ecology; Laboratory of Ecotoxicology LECOTOX

A direct discharge of untreated municipal wastewaters from the city of Novi Sad into the River Danube made a location a focal pollutant hotspot within the framework of FP 7 funded Solutions project. A study conducted previously at this site provided detailed chemical characterization of water samples. Relatively high concentrations of a number of chemicals were identified as of DRP toxicity was confirmed with a battery of in vitro assays. As a follow-up, a fish caging experiment was conducted with the aim to check if the biological effects observed in vitro can be seen in situ, through a battery of biomarkers in liver and brain of caged fish. Ten specimens of common carp (Cyprinus carpio L.), both male and female, were exposed for nine days at three sites in the River Danube: upstream, downstream and in the city center. Heart rate was the only downstream of the major discharge point of the untreated sewage into the River Danube. Certain detected chemicals are recognized causative agents of endocrine disruption and stress in general with a potential to lead to adverse physiological effects. Therefore, stress marker enzyme assays (catalase, carboxylesterase and glutathione t-transferase) were performed, and the expression of stress, endocrine disruption, immune response and autoantibody related genes was evaluated using qRT-PCR. Selected genes included cytochrome oxidase subunit 1 (cox1), metallothionein (mt), heat shock protein 70 (hsp70) as general stress related genes; estrogen receptor α (era), estrogen receptor β (erβ), androgen receptor (arr), cortical receptor (cr) and vitellogenin (vtg) as endocrine disruption related genes; interleukin1β (il1β) and tumor necrosis factor (tnf) as immune response related genes, while light chain 3 (β-lc3II) and dynen (dyen) were selected as autophagy related genes. Enzyme activities were higher in specimens caged at downstream locality, while catalase was lower at sewage discharge point. General stress and endocrine disruption representative genes at downstream site follow the trend of overexpression vs control (reference site), while the vsg was down-regulated at discharge point. Expression of mfn was strongly up-regulated at downstream site, while no significant difference was noted among expressions of other immune response and autophagy related genes. The results indicate induction of adaptive stress responses and endocrine disruption and are in line with the results observed in vitro. MO259 Gene transcription ontology of hypothalamic-pituitary-thyroid axis development in early-life stage fathead minnow and zebrafish


The hypothalamic-pituitary-thyroid (HPT) axis is known to play a crucial role in the development of teleost fish. However, knowledge of endogenous transcription profiles of thyroid-related genes in developing teleosts remains fragmented. We selected two model teleost species, the fathead minnow (Pimephales promelas) and the zebrafish (Danio rerio) to describe the gene transcription ontology of the HPT-axis. Control embryos were sampled at several time points between fertilization and hatching, and larvae were sampled approximately every other day until 33 days post-fertilization. Total RNA was extracted from pooled, whole fish, and thyroid-related mRNA expression was evaluated using quantitative polymerase chain reaction (qPCR) experiments. The study aims to define the role of certain genes and pathways during development. Furthermore, it can function as a background reference dataset for designing and interpreting targeted transcriptional expression studies both for fundamental research and for applications, such as ecotoxicology.
vtg-ir bands similar to those of females. Likewise, plasma E2 concentration in males was significantly increased but the opposite was observed in T levels. These changes were also reflected in E2/T ratio. Furthermore, E2 levels in treated males were even higher than those of females. Vtg and ERα gene expression was up-regulated both in liver and skin after E2 treatment. Potential impact in parental behavior is discussed. We conclude that vtg and ERα expression in skin are sensitive and non-harmful biomarkers of estrogenicity in this Sub-Antarctic fish.

MO261
Thyroid disruption and its effects on neuronal development of zebrafish
A. Hagis, Institute for Environmental Research, RWTH Aachen / Department of Ecosystem Analysis ESA; C. Di Paolo, RWTH Aachen University / Ecosystem Analysis ESA; L. Gebauer, RWTH Aachen University / Institute for Environmental Research
The endocrine disrupting effects of estrogenic compounds on humans and different species in the environment are well studied. But despite the growing numbers of patients diagnosed with thyroid disorders, thyroid hormone disrupting effects of compounds are less investigated. Furthermore, it is estimated that many substances found in our environment can influence the thyroid system and act as thyroid hormone disruptors. Thyroid hormones play a critical role in brain development and it has been shown that a lack reduces cognitive development. But the connections between thyroid disruption and developmental neurotoxicity are rarely studied and the basic mechanisms remain unknown. Because the thyroid system is well conserved among vertebrates, effects observed in humans can also be expected in wildlife. Out of several fish species that are of toxicological interest, zebrafish larvae are highly sensitive to toxicants. In this project an evaluation of the toxic effect of 2 pesticides: Dichlorvos which is an organophosphorus insecticide, used for the control of ectoparasites in fish, and insecticide Imiprotrin belonging to the group of pyrethroids, used as a main component in products for domestic use, was carried out. Because the previous studies with these products are scarce, the objective of this work was to evaluate its toxicity and its effects in 3 biomarkers: peroxidation of lipids (lipoperoxidation), the activity of the enzyme acetylcholinesterase (AChE) and the production of macromolecules (proteins, lipids and carbohydrates). Initially a test (96 hrs) was made, where zebrafish juveniles were exposed to 5 concentrations of pesticides (10, 1, 0.1, 0.01, 0.001, 0.0001 mg L\(^{-1}\)) to determine the 50 lethal concentration (LC\(50\)). Subsequently a bioassay with a duration of 15 days it was carried out where zebrafish juveniles were exposed to 2 sublethal concentrations (LC\(10\) and LC\(20\)). The results showed that AchE was decreased in both cases (-10 to -20%) than Dichlorvos (LC\(50\) = 5.3 mg L\(^{-1}\)). In the sublethal bioassays it was observed that the toxicity of these xenobiotics increased with the time of exposure. The degree of lipoperoxidation in the imiprotrin tests varied from 64.7 to 147.5 mN Tbars mg\(^{-1}\) and was higher than that observed in the bioassays with Dichlorvos (22.6 to 93.8 mN Tbars mg\(^{-1}\)). In the fish exposed to Dichlorvos a decrease of 43% to 86% in the activity of the AchE enzyme was observed and from 14% to 64% in the juveniles exposed to imiprotrin. The juveniles of zebrafish that showed a decrease in the activity of the AchE greater than 35% had changes in their swimming behavior and in their feeding. The energy content of the fish exposed to pesticides decreased by 64% in the Imiprotrin tests and 81% in the Dichlorvos bioassays. The insecticides Dichlorvos and Imiprotrin are little persistent in the environment, their half-life is 5 to 8 days, but the results of this study indicate that their effects on organisms are probably irreversible.

MO265
Effects of Omeprazole on zebrafish embryos (Danio rerio)
A. Sobrino-Figueroa, Universidad Autonoma Metropolitana Iztapalapa / Hidrobiología; J. Salazar Hernández, Universidad Autonoma Metropolitana Iztapalapa / Biología
Omeprazole is a proton pump inhibitor, it is used for the control of gastritis, for this reason it is one of the most prescribed drug, consumed by millions of people in the world. This drug has been associated with acid rebound hypersecretion, decreased growth, decreased brain metabolism and decreased lipid peroxidation. For this reason, effects of Omeprazole in the zebrafish has been studied in different species, but only in the larval stage. In this study an evaluation of the toxic effect of 2 pesticides: Dichlorvos which is an organophosphorus insecticide, used for the control of ectoparasites in fish, and insecticide Imiprotrin belonging to the group of pyrethroids, used as a main component in products for domestic use, was carried out. Because the previous studies with these products are scarce, the objective of this work was to evaluate its toxicity and its effects in 3 biomarkers: peroxidation of lipids (lipoperoxidation), the activity of the enzyme acetylcholinesterase (AChE) and the production of macromolecules (proteins, lipids and carbohydrates). Initially a test (96 hrs) was made, where zebrafish juveniles were exposed to 5 concentrations of pesticides (10, 1, 0.1, 0.01, 0.001, 0.0001 mg L\(^{-1}\)) to determine the 50 lethal concentration (LC\(50\)). Subsequently a bioassay with a duration of 15 days it was carried out where zebrafish juveniles were exposed to 2 sublethal concentrations (LC\(10\) and LC\(20\)). The results showed that AchE was decreased in both cases (-10 to -20%) than Dichlorvos (LC\(50\) = 5.3 mg L\(^{-1}\)). In the sublethal bioassays it was observed that the toxicity of these xenobiotics increased with the time of exposure. The degree of lipoperoxidation in the imiprotrin tests varied from 64.7 to 147.5 mN Tbars mg\(^{-1}\) and was higher than that observed in the bioassays with Dichlorvos (22.6 to 93.8 mN Tbars mg\(^{-1}\)). In the fish exposed to Dichlorvos a decrease of 43% to 86% in the activity of the AchE enzyme was observed and from 14% to 64% in the juveniles exposed to imiprotrin. The juveniles of zebrafish that showed a decrease in the activity of the AchE greater than 35% had changes in their swimming behavior and in their feeding. The energy content of the fish exposed to pesticides decreased by 64% in the Imiprotrin tests and 81% in the Dichlorvos bioassays. The insecticides Dichlorvos and Imiprotrin are little persistent in the environment, their half-life is 5 to 8 days, but the results of this study indicate that their effects on organisms are probably irreversible.
year. Considering the ecosystem services/principle, effects on single species, communities and whole ecosystems would increase that up to 'hundred times, similarly to when considering other chemicals such as neuroactive pharmaceuticals. 

\( \Delta \text{Antidepressants such venlafaxine are of increasing environmental neurotoxic concern. Venlafaxine is one of the most prescribed antidepressants in Europe and the U.S. and a known aquatic pollutant. } \frac{1}{2} \text{ is a serotonin-norepinephrine reuptake inhibitor, increasing serotonin and norepinephrine concentrations in brain regions. It was also shown to affect monoamine levels and cause behavioral alterations/inun fish. The aim of this study was to analyze the neurotoxic potential of Venlafaxine on zebrafish larvae by evaluating transcriptomic profiles and behavioral alterations. The locomotor activity in 'in vitro light-dark transition test and thigmotaxis were evaluated in 5 dpf larvae exposed to 24 h to 100 nM, 100 and 10 µM of venlafaxine using DanioVision}^{46} \text{ and EthoVision. A significant difference/inthe swimming behavior concerning the different concentrations could be detected. Effects on the transcriptome level were verified in zebrafish continuously exposed to Venlafaxine/1 nM, 100 nM up to 120 hpf. RNA was extracted from pooled samples (n = 25 fish) and submitted to} \frac{1}{2} \text{Sybr Green quantitative real-time chain reaction (qPCR). LCQ and pQCR were used to target gene selection and the considering targets involved in circadian rhythm regulation, muscle processes and responses/to abiotic stimulus. Behavioral results indicate decreased swimming distance and increased thigmotaxis/inun fish exposed, in agreement with previous own data for continuous venlafaxine exposure. Results/unin vitro qPCR indicated modulation of some of the pre-selected target genes such as sklp5, and currently 'unin confirmatory qPCR is being conducted. Further investigations in this system are planned to 0.1 µg/L were observed histological alterations in liver microstructure such as decrease of glycogen and progressive loss of hepatic architecture. The pattern of swimming behavior of fish changes significantly, fish spend more time at the upper part of the aquarium in concentration above 10 µg/L. Altogether, the present study demonstrated that chronic exposure of zebrafish to FLX can affect multiple endpoint such as growth tissue organization, feeding and swimming behaviour. These results emphasize the relevance of an integrated approach in the ecotoxicological assessment of psychiatric drugs.}

\section{MO267 \textbf{Acute effects of the ayahuasca infusion (Banisteriopsis caapi and Psychotria viridis) on zebrafish and rodent models}}

T.S. Andrade, Universidade de Brasilia / Laboratory of Genetics and Toxicology; W. Melo Junior, University of Brasilia; R. Oliveira, State University of Campinas / SCHOOL OF TECHNOLOGY - UNICAMP; A. D. ANDRADE, University of Brasilia / Laboratory of Toxicology, Faculty of Health Sciences; M.L. da Silva, University of Brasilia / genetic toxicology; J.A. Moraes, University of Brasilia / Laboratory of Toxicology, Faculty of Health Sciences; C. Koppe Grisolia, University of Brasilia / Department of Genetics and Morphology; I. Domingues, University of Aveiro / CESAM Department of Biology; E. D. Caldas, University of Brasilia / Laboratory of Toxicology, Faculty of Health Sciences; A. Pic-Taylor, University of Brasilia / Laboratory of Embryology and Developmental Biology; A. Vilaca, University of São Paulo / Laboratory of Toxicology, Faculty of Health Sciences; and A. S. Gouveia, USP / Laboratory of Toxicology, FHS. Ayahuasca is a psychoactive concoction prepared with the plants \textit{Banisteriopsis caapi} and \textit{Psychotria viridis} and used ancestrally by Amazonian Indian populations, and recently, by Christian religions in Brazil and other countries. The present study aimed at identifying the ayahuasca effects in early fish development and compares its neurobehavioral effects in the zebrafish embryo and rat models. Toxicity points for the zebrafish were analyzed at 0 and 1000 mg/L during 96 h of exposure. The effects on locomotor activity of zebrafish larvae were assessed using the automated video tracking system Zebraxbox at 0 to 20 mg/L and after 120 and 144 hours of exposure. The ayahuasca infusion was administered once by gavage to \textit{Wistar} rats at 1, 5 and 15 times the dose taken during a religious ritual, and neurobehavioral effects evaluated after 2 hours in the open field (OFT), elevated plus-maze (EPM) and forced swimming (FST) apparatus. The LC50 of ayahuasca in zebrafish was estimated to be 236.3 mg/L. Ayahuasca exposure caused significant developmental anomalies in zebrafish embryos, mainly at the highest concentration tested, including hatching delay, loss of equilibrium, edema and accumulation of red blood cells. The behavior of embryos was also significantly affected, with a decrease in locomotor activity at the highest tested concentration. Decreased locomotion was also observed in the rats treated at the highest dose in the OFT and EPM, and a higher swimming time in the FST, suggesting a possible antidepressant effect. These results indicated that the effects of ayahuasca correlated well for zebrafish embryos and rodents, showing that zebrafish may provide a useful model to study ayahuasca and other hallucinogenic drugs. A research focusing on the molecular pathways affected by ayahuasca administration in both zebrafish and rat models could provide additional information on the potential of ayahuasca as an antidepressant. Acute effects of the ayahuasca infusion (\textit{Banisteriopsis caapi} and \textit{Psychotria viridis}) on zebrafish and rodent models

\section{MO268 \textbf{Chronic exposure to fluoxetine affects growth, feeding, swimming behavior and tissue organization of zebrafish.}}

N. de Farias, University of Brasilia / Departamento de Genética e Morfologia; R. Oliveira, State University of Campinas / Department of Genetics and Morphology; T.S. Andrade, Universidade de Brasilia / Laboratory of Genetics and Toxicology; J.M. Pinto, University of Brasilia / Departamento de Genética e Morfologia Instituto de Biologia; C.K. Grisolia, University of Brasilia UNB / Department of Genetics and Morphology. Fluoxetine (FLX) is among the top 100 drugs prescribed annually worldwide. This selective serotonin reuptake inhibitor is highly detected in aquatic ecosystems and has the potential to modulate levels of serotonin of non-target organisms. The present study aims to evaluate the effects of chronic exposure to FLX on fish. Zebrafish juvenile were exposed to FLX during 30 days following the OECD protocol (no. 215). The assays were performed in triplicate with six treatments of FLX: 0; 0.01; 0.1; 1; 10 and 100 µg/L. A total of 60 fish per treatment (20 per replicate) were used. Growth and feeding behaviour were analysed at the end of the test. Histological analyses of liver followed standard H&E routine. Video recordings (10 min) were analysed for swimming behaviour by measuring the time spent by each fish in each of the layers of the aquarium (bottom: 0–5 cm, middle: 6–10 cm and upper: 11–15 cm). Our results showed decrease in growth rate and erratic feeding behaviour at 100 µg/L. Also, in concentrations as low as 0.1 µg/L were observed developmental alterations such as microcephaly. These results are compared with the activity of mitochondrial complex I–IV after isolating mitochondria from embryos at 48 hpf. In addition, we analyzed mRNA expression of transcription factors (i.e., \textit{PGC-alpha}, \textit{Acoc1}, \textit{SDHA}, \textit{MCAD}, and \textit{CS}), associated with mitochondrial metabolism, at 120 hpf. This comprehensive results could suggest the effects of the embryonic zebrafish model on the methodology and a set of research scheme to determine mitochondrial disorders in the exposure of individual OCPs and their mixtures.

\section{MO269 \textbf{Mitochondrial Disorders of Zebrafish Embryos Exposed to Individual Organochlorine Pesticides and Their Mixtures}}

J. Lee, Seoul National University of Science and Technology / Environmental toxicology and health; S. Lee, Seoul National University of Science and Technology / Environmental engineering; K. Kim, Seoul National University of Science and Technology / Environmental Engineering. Organochlorine pesticides (OCPs), prohibited compounds in the 1970s, are still being detected in human and environmental samples. Mitochondrial dysfunction caused by chemical exposure has attracted great attention on toxicological studies. We evaluated mitochondrial dysfunction in dechorinated zebrafish embryos exposed to individual 5 OCPs (i.e., \textit{p,p-DDT}, Chlordane (mixture), Heptachlor, Hexachlorobenzene (HCB), and beta-hexachlorocyclohexane (beta-HCH)), and their mixtures from 4 to 120 hpf (hours post-fertilization). We measured oxygen consumption rate (OCR) at the embryonic sublethal concentrations of 0.05, 0.1, and 0.5 mg/L by using Seahorse XF Extracellular Flux Analyzer at 24 hpf. The OCR results are compared with the activity of mitochondrial complex I–IV after isolating mitochondria from embryos at 48 hpf. In addition, we analyzed mRNA expression of transcription factors (i.e., \textit{PGC-alpha}, \textit{Acoc1}, \textit{SDHA}, \textit{MCAD}, and \textit{CS}), associated with mitochondrial metabolism, at 120 hpf. This comprehensive results could suggest the effects of the embryonic zebrafish model on the methodology and a set of research scheme to determine mitochondrial disorders in the exposure of individual OCPs and their mixtures.

\section{MO270 \textbf{The NeuroBox Project}}

H. de Haas, Vrije Universiteit Amsterdam; A. Haigis, Institute for Environmental Research RWTH Aachen University; M. Gundlach, RWTH Aachen University / Department of Ecosystem Analysis (ESA); C. Di Paolo, RWTH Aachen University / Ecosystem Analysis (ESA); M. Spehr, B. Kampa, RWTH Aachen University; H. Hollett, RWTH Aachen University / Institute for Environmental Research. The societal impact of neurological disorders like Alzheimer’s disease or neurodevelopmental defects like autism is immense. To better understand these diseases, the effects, e.g. severe mental and physical problems, are often devastating. There is mostly no cure available and even treatments to reduce or stop the progression of the diseases are limited. The number of people diagnosed with neurological disorders is increasing. This increase cannot be explained by improved diagnostics and increased age. Exposure to neurotoxic chemicals is suspected to play a role in the development and progression of these diseases. It has been estimated that alone in Europe, exposure to solely endocrine disrupters that lead to neurological disorders, costs society €150 billion per year. This does not include costs due to exposure to known neuroactive substances like pesticides and pharmaceuticals. Currently, testing for neurotoxicity is not required within the EU (REACH) as it is not yet clear how to assess these effects. Considering the large number of chemicals and the physiological and morphological complexity of the nervous system, it is a major challenge to test all substances for their neurotoxic potential, new advanced neurotoxicity assessment strategies need to be developed to fulfil these demands. The bmbf funded project NeuroBox (02WR1S1419; coordination UBA, T. Grummt) aims to develop novel assessment strategies for neurotoxicity assessment of anthropogenic substances in water samples. The work is split over six subprojects. In our sub-project, we use zebrafish embryos to identify neurotoxic modes of action of commonly found water contaminants. Based on our findings novel screening assays will be developed to easily screen water samples for neurotoxic effects. In combination with mouse models and clinical researchers we will also link our results to mammalian neurological diseases like ADHD and autism and neurodegeneration studies, to identify potential exposure–disease relationships. Furthermore, we will study the link between endocrine disrupting compounds and neurotoxic effects. We could see behavioral as well as metabolomics and transcriptomic changes after exposure to compounds. These
changes were observed at concentrations bellow any phenotypic changes could be observed. Our results so far show that assessing neurotoxicity is complex and a tiered approach covering behavioral tests in combination with OMICS techniques seem to be a cost and time efficient way.

MO271
Understanding the correlation between behavioural inter-individual variability and physiology/morphology in zebrafish larvae
K.T. Kiria, C.M. von Berg, Eawag Swiss Federal Institute of Aquatic Science and Technology / Environmental Toxicology

Background: Zebrafish are widely used in biomedical research as they offer several features such as the fully sequenced genome, availability of a wide range of mutants, generation of large numbers of offspring and the possibility to raise and conserve vertebrate drug targets to a great extent. Early stages of zebrafish are called as non-protected life stages and are therefore used for testing chemicals for their toxicity as an alternative to conventional animal testing. Moreover, zebrafish larvae are amenable to test neurotoxicity and behavioural effects of chemicals as their small body size allows tracking a large number of individuals with full control over the environment. Aim: The aim of the project is to investigate behavioural inter- and intra-individual variability in zebrafish larvae as a basis to better estimate effects of chemicals on behavioural responses. Analysis of inter-individual differences might offer new insights into mechanisms of toxicity considering that every individual’s response to a chemical differs based on their genetic make-up.

Hypothesis: We are testing whether inter-individual variability is constant over time and whether levels of locomotor activity correlate with physiological and morphological properties of the larvae. Methods: At first, spontaneous locomotor activity is measured for 40 min in continuous light at different timings of the day from 5-7 days post fertilization. Heart rate, body size and other physiological properties of the same individuals are analysed at different time points. Results: From the preliminary results of the locomotor activity analysis, we could assign the larvae to three categories based on their activity levels compared to the average activity: highly active, less active and the individuals close to the average activity, which are also the ones less variable over time. To attribute this variability in the individual’s activity to its physiology and phenotype, the analysis of heart rate, length and blood flow are on-going. Outlook: The variability of each individual will be taken into account to better evaluate effects of the chemicals on behavioural responses. Inter-individual differences will be explored as a source of information on mechanisms of toxicity of chemicals with unknown targets and mode of action.

MO272
Effect of iodinated X-ray contrast media in the formation of disinfection byproducts during chlorination and chloramination of water
C. Postigo, IDAEA, CID-CSIC / Environmental Chemistry; M. Armstrong, University of North Carolina at Chapel Hill / Department of Environmental Science and Engineering; C. Herb, University of Akron; S. Duirk, University of Akron; J. Simmons, U.S. Environmental Protection Agency / National Health and Environmental Effects Research Laboratory; S. Duirk, University of Akron; J. Simmons, U.S. Environmental Protection Agency / National Health and Environmental Effects Research Laboratory; C. Postigo, IDAEA, CID-CSIC / Environmental Chemistry.

Iodinated X-ray contrast media (ICMs), used in medical imaging, are poorly metabolized by humans and enter wastewater. As they are incompletely removed during wastewater treatment, ICMs are released to the aquatic environment and have been detected in drinking water sources. ICMs have been identified as iodine byproducts during wastewater treatment, ICMs are released to the aquatic environment and are a cost and time efficient way.

Environmental fate of emerging contaminants in the water cycle: analytical challenges and engineered solutions (P)

MO274
Balancing environmental quality standards and infrastructure upgrading costs for the reduction of microcontaminants loads in rivers
V. P. Simonetto, CIAD, Catalan Institute for Water Research (ICRA); J. SEVERYNS, AQUAFIN; J. Comas, L. Coronas, Catalan Institute for Water Research ICRA Investments for upgrading wastewater treatment plants (WWTPs) with tertiary treatment to reduce microcontaminants loads in surface waters at a catchment scale can be daunting. Our hypothesis was that these investments seriously change upon selection of the Environmental Quality Standards (EQS) for unregulated microcontaminants, and hence there is a trade-off between EQS selection and investment which needs to be considered in decision-making. We used a customized Microcontaminant Fate and Transport Model coupled to an optimization algorithm to validate this hypothesis. We used the Llobregat river basin as a case study and dicrofenc as the unregulated microcontaminant. The algorithm optimized the selection of WWTPs in this catchment requiring an upgrade to minimize the EQS exceedance of dicofenc in total chemical load. We simulated 40 scenarios representing a combination of 4 potential EQS which are currently being discussed in the European Union (10, 30, 50 and 100 ng/l), 5 levels of uncertainty bounds in the predictions of river concentrations and 2 hydrological scenarios (average flows and low flows). The results showed that the operation of WWTPs for tertiary treatment in the Llobregat river basin was 6 M€/year (upgrading 8 WWTPs out of the existing 56 for fulfilling an EQS of 30 ng/l in the entire catchment). Such an investment seriously changed upon selection of EQS. The cost varied from 6 M€/year (upgrading 3 WWTPs for fulfilling an EQS of 100 ng/l) to 13 M€/year (upgrading 18 WWTPs, for fullfilling an EQS of 10 ng/l). We observed that the selection of catchment hydrological scenarios during the upgrading analysis also plays a key role. The cost of the upgrades when considering low surface water flows (minimum environmental flows that ensure compatibility of environmental needs and human water consumption) was 50% higher than the cost obtained with average flows (average 201

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hydrological conditions in the Llobregat). Finally, we demonstrated that the reduction of uncertainty in the modelling process (through R&D activities) provides transparency in the decision-making process.  

**MO275**  
Calibration of passive samplers for the monitoring of drugs in French Caribbean.  
N. Tapie, Univ. Bordeaux, CNRS, EPOC UMR 5805 / EPOC UMR 5805; D. Devault, Univ. Paris Sud / ESE UMR 8079; S. Karolak, Univ. Paris Sud, CNRS, AgroParisTech / ESE UMR 8079; Y. Levi, Univ. Paris Sud / ESE UMR 8079; H. Budzinski, University of Bordeaux  

Drugs are more and more consumed worldwide (ONUDC, 2017). The French Caribbean is a hub of world cocaine trafficking and an important place of consumption of cocaine in the form of crack. The local population is particularly affected by this scourge. The consumption of illicit drugs induces the excretion of parent compounds or metabolites (markers of drugs uses) in waste water, and in the end their release into the aquatic environment. So, one of the ways to evaluate the local use of illicit drugs is to track drug residues in waste water treatment plants (WWTP). The present study that takes place in the SENEUR Project and explores the use of passive sampling techniques to monitor illicit drugs in WWTP. Polar Organic Chemical Integrative Samplers (POCIS) were exposed in situ in a Waste Water Treatment Plant in Martinique (French Caribbean) during ten days. First an analytical development by ESI-LC/MS/MS was done in order to be able to analyze 17 compounds as markers of drug uses (cocaïne, heroin, amphetamine, cannabis, their main metabolites and some substitute products such as methadone) in effluent and in POCIS (LOQ from 0.01 to 0.1 pg/g). Secondly triplicates of POCIS were exposed in WWTP for calibration during 10 days. POCIS were collected at different times T0 days, T1 days, T2 days, T3 days, T4 days, T5 days, T6 days, T7 days, T8 days, T9 days, T10 days. Water samples were also daily collected. The first result of the calibration show a good capacity of the POCIS to sample cocaine markers (cocaïne, benzoylcegonine, cocaethylene, cocaetanol) and cannabis markers (vivian, THC, THCA). The concentrations were also determined through a spiking test to check the selectivity of the POCIS during the measurement. To monitor the immission situation in 15 surface and urban water bodies with various matrices. These workflows in a form of general steps can be extrapolated to other catchment. The plotting of other compounds concentrations >150 ng/L perchlorate, and accuracy, was 95.6-102% for concentrations >150 ng/L perchlorate, and 111% for concentrations < 150 ng/L perchlorate. Single laboratory precision in high-ionic-strength matrix, was < 5% at concentrations >150 ng/L perchlorate, and accuracy, was 100-103.5% for concentrations >150 ng/L perchlorate.  

**MO276**  
Passive sampling in surface water as an immision-based approach to extrapolate waste-water-related pressures and potential EQS exceedence in Luxembourg.  
T. Galle, Luxembourg Institute of Science and Technology; D. Pittois, M. Bauerle, Luxembourg Institute of Science and Technology LIST  

The pressure on water surfaces that is exerted by emerging pollutants depends on the WWTPs treatment capacity and the quality of the effluent. Both can be variable depending on the compound, the contributors of the sewer network as well as the design and operation of the treatment plant. Several emerging compounds have mixed uses and can therefore stem from domestic as well as from industrial sources. Regulators have an interest in knowing immission situations that will probably lead to EQS exceedences without needing to monitor emerging pollutants in the whole catchment. The objective was to develop a non targeted approach to monitor the immission situation in 15 surface waters under low-flow conditions and different sanitary pressures in Luxembourg. We define sanitary pressure as the number of population equivalents divided by the surface of the catchment for a monitoring point. For that purpose, a catchment delineation is performed for each measurement point and the PE of discharging treatment plants within the catchment is determined. The approach is based on the detection of markers by natural flow. Since WWTPs are relatively constant sources sampling rates of passive samplers can easily be calibrated with grab samples over all monitoring locations. The data evaluation uses the conservative behaviour of carbamazepine as a tracer for (treated) wastewater input. Carbamazepine concentrations proved to be correlated to the sanitary pressure (PE/ha) in a catchment. The plotting of other compound concentrations against carbamazepine holds useful information: it shows the variability of the WWTP influents as well as elimination capacities in the catchment. According to these hypotheses recalcitrant pharmaceuticals showed very strong and narrow linear correlations with carbamazepine while intermediate- degradable compounds displayed higher variability. Complete outliers make it easy to detect industrial sources as was the case for triazoles for instance. Finally the data set made it possible to extrapolate expected concentrations of emerging compounds for different sanitary pressure levels and by integrating EQS values, to define a threshold of 2.5 PE/ha above which EQS exceedance for diclofenac and clarithromycine is expected. This makes it easy to design a map of the river network with segments at risk with basic population equivalent information.  

**MO277**  
Determination of Perchlorate by U.S. EPA Method 332.0 Using a Compact Ion Chromatography System Coupled with Mass Spectrometry (IC-MS)  
B. Huang, Thermo Fisher Scientific / marketing; T. Cross, Thermo Fisher Scientific; J. Rohrer, Thermo Fisher Scientific / Chromatography and mass spectrometry division  

Perchlorate has been used as an oxidizer in rockets, munitions, and fireworks since the 1950s. It has been found to cause thyroid dysfunction, and has been linked to breast tumors in humans. Perchlorate is regulated under the Safe Drinking Water Act (2011). Massachusetts and California have established standards for drinking water of 2 μg/L and 6 μg/L, respectively. Determination of perchlorate in environmental samples has also gained the attention of the International Standards Organization (ISO). U.S. EPA Method 332.0 — Ion Chromatography with Suppressed Conductivity and Electrospray Ionization/Mass Spectrometry is one of the most sensitive and robust characterization methods available for perchlorate determination (MS) providing detection limits in high-ionic-strength matrices with conductivity detection alone. These low detection limits are achieved without sample preparation. Our study updates the IC-MS method published in U.S. EPA Method 332.0 for determination of perchlorate in environmental waters. The method uses a Thermo Scientific™ Dionex™ IonPac™ AS20 column set, on a recently introduced compact IC system coupled with a recently introduced single quad mass spectrometer. The selectivity of the mass spectrometer allows the quantification of perchlorate in high-ionic-strength samples at well below currently enforced action levels. Ionization improvements to the electrospray source eliminate the need to add organic solvent to enhance detection. Method detection limit (MDL) values in deionized water are 20- 60 ng/L, and MDLs in high-ionic-strength matrix are 30- 60 ng/L. The calibration curves for perchlorate in high-ionic-strength matrix at 101 μg/L over the range of 125-5000 ng/L using the internal standard and external methods showed good linearity with the coefficient of determination being 0.9993, and 0.9998 respectively. Single laboratory precision in drinking waters, as measured by RSD, was < 5% at concentrations >150 ng/L perchlorate, and accuracy, was 95.6-102% for concentrations >150 ng/L perchlorate, and 111% for concentrations < 150 ng/L perchlorate. Single laboratory precision in high-ionic-strength matrix, was < 5% at concentrations >150 ng/L perchlorate, and accuracy, was 100-103.5% for concentrations >150 ng/L perchlorate.  

**MO278**  
NEW OPPORTUNITIES FOR THE NON TARGETED ANALYSIS OF ENVIRONMENTAL CONTAMINANTS USING GAS CHROMATOGRAPHY- ORBITRAP MASS SPECTROMETRY  
P. Silcock, Thermo Fisher Scientific / GC-MS; D. Cardona, Thermo Fisher Scientific / Environmental Analysis  
Since the middle of the 20th century GC-MS has made a long journey towards its current status as one of the major analytical techniques used in a diverse range of applications. Despite this, GC-MS has had more than four decades to wait for a new type of mass analyzer with the potential to advance capability over previously applied technology. Almost two years on from the first commercial introduction of Orbitrap GC-MS in 2015, in this presentation, we explore how this technology has been applied specifically to the analysis of environmental contaminants and how non targeted acquisition can be used as a tool for the development of a new approach to routine environmental analysis. Primary applications to be highlighted are the discovery of new disinfection by-products (DBPs) resulting from water treatment processes, using a non targeted approach, as well as the potential for addressing the difficult analytical challenges for a complex class of emerging persistent organic pollutants: short chain chlorinated paraffins (SCCPs).  

**MO279**  
HILIC workflow strategy for the hidden target screening of very polar compounds in surface waters  
S. Veloutous, Technical University of Munich; S. Bieger, Technical University of Munich / Chair of Water Systems Engineering; S. Grosse, T. Letzel, Technical University of Munich / Chair of Water Systems Engineering  

Trace Organic compounds (ToOCs) in water can be biogenic or anthropogenic. These compounds can range a lot in terms of polarity. Reverse Phase Liquid Chromatography (RPLC) is the most common and widely used tool for the separation of non-polar and mildly polar compounds. However, for the separation of very polar compounds, techniques like the Hydrophilic Interaction Liquid Chromatography (HILIC) are needed. HILIC has been established since years as an analytical tool, capable to separate effectively very polar molecules. Using a serial RPLC-HILIC system coupled with ToF-MS the analytical screening of samples comprised of solutes with variability in structure and polarity can be achieved. Full-spectrum acquisitions in non-target screening approaches are producing large datasets with the detected features of the samples. Different workflows have been published, proposing ways to cope with the collected amount of data in an automatic, time efficient and reproducible way, which can be applied to samples with various matrices. These workflows in a form of general steps can be summarized as: a) filtering and prioritizing the detected features (peak picking), b) molecular formula assignment, and c) a search in one or more compound databases. A newly active young compound database for water relevant compounds is STOOF-IDENT. In order to achieve a comprehensive identification of the water’s organic content, Non-target screening strategies have become increasingly popular. This study was realized by analyzing river water samples with the established RPLC-HILIC-ToF/MS system and by using the STOOF-IDENT compound database. Its main aim is to demonstrate and discuss an efficient strategy for the non-target screening of aqueous environments, as a mean to facilitate the process of identification of very polar compounds. Three 24h composite samples were collected using time proportional samplers; upstream and downstream of a WWTP and wastewater effluent. The samples were analysed by the established
RPLC-HILIC-ToF/MS system. The analysis data were then processed following a non-target screening workflow for very polar compounds. After importing the data in STOFF-IDENT database, a proposed list of possible compounds in the samples was created. Using reference standards of the proposed compounds and MS/MS fragmentation data, it was possible to positively identify nine very polar compounds, of which six have not been reported previously in water surface samples.

MO28
Analysis of Per/Polyfluoroalkyl Substances (PFAS) in Drinking Water using LC/MS/MS to meet USEPA 537 requirements
T. Anamol, L. Toelwezi, T. Sosienski, Agilent Technologies; D. Cardona, Climate Change Canada; A. Holscher, Environment and Climate Change Canada / Aquatic Contaminants Research Division; S. Nzube, Cape Peninsula University of Technology / Department of Chemistry; C. Van der Horst, University of The Western Cape / SensorLab Department of Chemistry; V. S. Somerset, CPUT / Chemistry

Per/Polyfluoroalkyl Substances (PFASs) are organic molecules that have a C-F bonds in the alkyl chain and have a range of industrial uses and can be found in various household items and consumer goods. PFASs however can have adverse health effects while longer (C-chain >7) PFASs are bioaccumulative. PFASs are extremely persistent and have been detected in various environmental matrices. Consequently, the USEPA has public health guidelines in drinking water for two PFASs, namely perfluorooctanoic acid (PFOA) and perfluorooctanesulfonic acid (PFOS) at 70 ng/L. However, several other PFASs are also used in manufacturing and need to be monitored. To respond to this, a method was developed for the detection of PFASs in drinking water using a newly developed triple-quadrupole mass spectrometer and a modified high-performance liquid chromatography system. The USEPA has developed a method for analysis in drinking water for 14 PFAS and this method expands on that method with lower detection limits, and more QA/QC data. The water samples were extracted with solid phase extraction using a novel weak anion exchange cartridge which was optimized to achieve good recoveries for all compounds and will be presented. Several different classes of PFASs including perfluorocarboxylic acids (PFCAs), perfluorosulfonic acids (PFSA)s, sulfonamides (FOSA), sulfonamide acetic acids (FOSAs) and others were separated on a liquid chromatography (LC) using a reversed phase C-18 column. Since fluoropolymers are used in all LC systems, special precautions including replacing solvent lines and addition of a delay column were employed to avoid PFAS background contamination. The compounds were analyzed in negative electrospray ionization using a tandem quadrupole mass spectrometer in dynamic multiple reaction monitoring (MRM) mode. Water samples were extracted using both an offline and automated online solid phase extraction techniques and the data was compared. All PFASs were analyzed and method performance parameters such as method detection limits, inter- and intra-day repeatability, matrix spike recoveries and other QA/QC criteria were evaluated. All recoveries were with 70-125% with %RSDs well below 15% that are needed to meet USEPA 537 requirements. Such a method for the detection of PFOS and PFOA in drinking water and wastewater samples that were analyzed in the ng/L range. All this was done on a triple quadrupole mass spectrometer that is fully stackable with the HPLC system.

MO282
Optimisation of solid phase extraction parameters for the isolation and characterisation of benzodiazepines in wastewater
S. Nzube, Cape Peninsula University of Technology / Department of Chemistry; C. Van der Horst, University of The Western Cape / SensorLab Department of Chemistry; V. S. Somerset, CPUT / Chemistry

Pharmaceutical pollutants entering the aquatic environment have become a growing concern the last decades. These pharmaceuticals are unique pollutants because of their special characteristics and behaviour that cannot be simulated with other organic pollutants. The untreated wastewater effluent that contains pharmaceuticals poses a considerable threat to the aquatic ecosystem because of the negative effects of non-target organisms in the water. Recent years have seen a growing concern about the benzodiazepines, as emerging pollutants, and their effects on the aquatic environment. These compounds are nowadays widely detected in sewage wastewater. It is important to increase the emphasis on the characteristics of the benzodiazepines in order to differentiate them from industrial chemical contaminants. In this study, various solid phase extraction techniques have been employed focussing on isolation of benzodiazepines in wastewater matrices. Emphasis was put on the development of different SPE/SFE techniques to enrich cephamycin and lorezapam as benzodiazepines. Keywords: Benzodiazepines; Emerging pollutants; Solid Phase Extraction; Spectroscopy; Wastewater

MO283
Monitoring source and drinking waters for Microcystins using online LC/MS/MS method
J. Westrick, Wayne State University / Lumigen Instrument Center; D. Cardona, Thermo Fisher Scientific / Environmental Analysis

In 2015 the USEPA announced an age-dependent drinking water Health Advisory (dWA) for the natural freshwater toxins, microcystins (MCs). For pre-school age children and adults, the MC dWA values are 0.3 mg/L and 1.6 mg/L, respectively. Although the dWA values are non-regulatory values, this announcement compels compelling health information that cannot be ignored. In parallel, EPA Method 544, a solid phase extraction/liquid chromatography tandem mass spectrometry (LC/MS/MS) method was released. Our goal was to create an online concentration LC/MS/MS method with 12 MCs that meets the EPA’s quality assurance/quality control (QA/QC) criteria. MC concentrations were measured in samples from freshwater lakes and drinking water. Samples were prepared by three freeze/thaw cycles, centrifuging, and filtering through a 0.25 μm polycarbonate filter. Our LC/MS/MS platform included an online sample concentrator with UHPLC for separation and a triple quadrupole mass spec for MS/MS analysis. This method included 12 MCs with IC50 concentration curves from 0.5 – 500 μg/L with values greater than 0.996. The MCs eluted between 2.2 – 5.2 minutes allowing for the analyses time to be 3) MC-RR, [Asp] MC- LR, MC-HiR, and MC-WR at concentrations above the low health reference level of 21 ng/L. Our data shows that 1) by not including 12 MCs in Method 544, the true risk potential of exposure to MCs in drinking and recreational waters will be underestimated greatly, and 2) an automated online system needs to be employed for this application. Finally, our LC/MS/MS method reduces sample preparation, chemical usage, and instrument and preparation time while meeting EPA quality assurance criteria.

MO284
Prioritising site-specific emerging contaminants in surface water based on LC-HRMS non-target screening data
M. Krauss, C. Hug, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis; R. Bloch, Helmholtz Centre for Environmental Research GmbH / UFZ; T. Schulze, W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis

Fish and seafood contaminant monitoring traditionally relies on the surveillance of known chemicals, an approach referred to as targeted analysis. However, very few tools are available to monitor “unexpected” or “unknown” compounds. Non-targeted approaches are particularly useful to detect emerging contaminants in items related to the human diet such as fish and seafood. The non-targeted approach is however quite challenging for trace contaminant analysis as it involves isolating relatively small signals from complex matrices, and this, in absence of a good knowledge about the analyte identity. In the past decade, the coupling of liquid chromatography, high-resolution tandem mass spectrometry (HRMS/MS) and advanced data processing algorithms has proved to be a robust approach for the analysis of unknown molecules in biological samples. In this study, a non-targeted workflow was developed with the objective to determine/identify unexpected organic contaminants in a predator fish from the St. Lawrence River (QC, Canada), the northern pike (Esox lucius), with a focus on chemicals originating from plastic materials. An optimized method was applied to pike tissue sampled upstream and downstream of the Montreal’s wastewater treatment plant. The two sampling sites (upstream vs downstream) were then compared using Mass Profiler Professional Software for the presence of other unexpected contaminants. The final confirmation of various substances of interest (e.g. PFOS) was investigated through the comparison with analytical standards. Results indicated that the non-targeted workflow optimized in this study can successfully identify unexpected chemical residues in fish matrices.

MO285
Development of a LC-MS/MS-based method for the screening of non-targeted chemicals of potential concern in northern pike
L. Tian, McGill University; J. Reinling, Université du Québec à Montréal / Département des sciences biologiques; J. Verreault, Université du Québec à Montréal / Département de Biological Sciences; M. Houde, Environment and Climate Change Canada / Aquatic Contaminants Research Division; S. Bayen, McGill University / Singapore-Delft Water Alliance

Finally, our LC/MS/MS method reduces sample preparation, chemical usage, and instrument and preparation time while meeting EPA quality assurance criteria.

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Non-targeted approaches are particularly useful to detect emerging contaminants in items related to the human diet such as fish and seafood. The non-targeted approach is however quite challenging for trace contaminant analysis as it involves isolating relatively small signals from complex matrices, and this, in absence of a good knowledge about the analyte identity. In the past decade, the coupling of liquid chromatography, high-resolution tandem mass spectrometry (HRMS/MS) and advanced data processing algorithms has proved to be a robust approach for the analysis of unknown molecules in biological samples. In this study, a non-targeted workflow was developed with the objective to determine/identify unexpected organic contaminants in a predator fish from the St. Lawrence River (QC, Canada), the northern pike (Esox lucius), with a focus on chemicals originating from plastic materials. An optimized method was applied to pike tissue sampled upstream and downstream of the Montreal’s wastewater treatment plant. The two sampling sites (upstream vs downstream) were then compared using Mass Profiler Professional Software for the presence of other unexpected contaminants. The final confirmation of various substances of interest (e.g. PFOS) was investigated through the comparison with analytical standards. Results indicated that the non-targeted workflow optimized in this study can successfully identify unexpected chemical residues in fish matrices.

MO286
Directed Analysis of Per/Polyfluoroalkyl Substance (PFAS) in Drinking Water using LC/MS/MS to meet USEPA 537 requirements
M. Krauss, C. Hug, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis; R. Bloch, Helmholtz Centre for Environmental Research GmbH / UFZ; T. Schulze, W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis

Finally, our LC/MS/MS method reduces sample preparation, chemical usage, and instrument and preparation time while meeting EPA quality assurance criteria.
Annotation. These sulfur-containing compounds could be identified as various derivatives of naphthalene sulfonic acids and have to be considered as site-specific contaminants, as they were not present at any other sampling site. Thus, the proposed approach is suitable to rapidly characterize surface water samples and allows for a prioritization of sites or compound groups for further in-depth studies.

MO286
Analysis of Phenanthrene Transformation Products Using High-Resolution Mass Spectrometry Coupled to High-Performance Liquid Chromatography M. Leonard, Oregon State University / Environmental & Molecular Toxicology; J. Schrlau, Oregon State University / Environmental and Molecular Toxicology; S.L. Moorey Simonow, Oregon State University / Department of Environmental and Molecular Toxicology Polycyclic aromatic hydrocarbons (PAHs) are environmental contaminants generated by the incomplete combustion of organic compounds. Several PAHs have been identified as toxic, mutagenic and/or carcinogenic, which has led the United States Environmental Protection Agency (US EPA) to list 16 PAHs as a priority pollutants. PAHs can be metabolized by mammals and some microbes to form hydroxylated PAHs (OHPAHs) and a variety of other transformation products (TPs). These TPs have the potential to be more toxic than their parent PAHs, but they are not included on the EPA priority pollutant list. Hence, they are often not screened for in environmental samples. Non-targeted screening based on high-resolution mass spectrometry (HRMS) coupled to high-performance liquid chromatography (HPLC) has been shown to be a useful approach to address this issue. In this study, we developed a method to analyze phenanthrene TPs detected in bioremediated water. C10, phenyl-hexyl, and fluoro-phenyl HPLC columns were evaluated for their ability to resolve hydroxypheanthrene (OH-Phe) isomers. Baseline resolution of 2, 4-, and 9-OH-Phe was achieved with the C10 and phenyl-hexyl columns using a gradient of water:mobile phase A) and a mixture of acetonitrile and methanol (mobile phase B). Gradient elution beginning with a relatively high percentage of organic solvent (≥ 70%) yielded satisfactory separation and peak shape without the use of an additive or buffer. Detection was carried out with a high-resolution time-of-flight MS employing electrospray ionization (ESI) in negative ion mode. The high organic solvent composition of the eluent enabled optimal ESI performance. Consequently, sensometric sensitivity was preserved throughout each analysis. Further investigation will determine whether the fluoro-phenyl column is suitable for separation of OH-Phe isomers. The method will be used for separation of phenanthrene metabolites and other PAH TPs in non-targeted screening of bioremediated mixtures.

MO287
Strategies to monitor transformation products in the water cycle A. Brunner, D. Vughs, W. Siegers, C. BertylKamp, R. Hofman-Caris, A. Kollmann, S. Kools, T. ter Laak, KWR Water cycle Research Institute Transformation products (TPs) are formed in the water cycle through both biotic and biotechnological processes. Data availability showed that TPs in complex environmental samples. An HPLC-HRMS method was developed for analysis of phenanthrene TPs detected in bioremediated water. C10, phenyl-hexyl, and fluoro-phenyl HPLC columns were evaluated for their ability to resolve hydroxypheanthrene (OH-Phe) isomers. Baseline resolution of 2, 4-, and 9-OH-Phe was achieved with the C10 and phenyl-hexyl columns using a gradient of water:mobile phase A) and a mixture of acetonitrile and methanol (mobile phase B). Gradient elution beginning with a relatively high percentage of organic solvent (≥ 70%) yielded satisfactory separation and peak shape without the use of an additive or buffer. Detection was carried out with a high-resolution time-of-flight MS employing electrospray ionization (ESI) in negative ion mode. The high organic solvent composition of the eluent enabled optimal ESI performance. Consequently, sensometric sensitivity was preserved throughout each analysis. Further investigation will determine whether the fluoro-phenyl column is suitable for separation of OH-Phe isomers. The method will be used for separation of phenanthrene metabolites and other PAH TPs in non-targeted screening of bioremediated mixtures.

MO289
Unravelling the potential of a partial nitritation/anammox biomass towards micropollutants biodegradation E. KASSOTAKI, ICRA (Catalan Institute for Water Research) / Technologies and Evaluation; M. PIJAN, Catalan Institute for Water Research ICRA / Technologies and Evaluation; L. GASMARSI, Catalan Institute for Water Research ICRA; I. RODRIGUEZ RODA, Universitat de Girona and ICRA / LEQUA/IA; A. Joss, Eawag Swiss Federal Institute of Aquatic Science and Technology; G. Buttigliere, Catalan Institute for Water Research ICRA In the past few years, anammox-based processes have attracted a lot of attention for their implementation at the mainstream line of wastewater treatment plants, due to the possibility of leading to energy autarky if combined with anaerobic digestion. However, little is known about the potential degradation of micropollutants by the microbial groups responsible of these processes and the few results available are inconclusive. This study aims to assess the degradation capability of biomass withdrawn from a partial nitritation/anoxic ammonium oxidation (PN/A) pilot plant towards five pharmaceutically active compounds (ibuprofen, sulfamethoxazole, metoprolol, venlafaxine and carbamazepine). Batch experiments were performed under different conditions by selecting different microbial groups: i) regular PN/A operation, ii) aerobic (optimal for nitrifying bacteria), iii) aerobic conditions with allylthiourea (an inhibitor of ammonia oxidizing bacteria), iv) anoxic (optimal for anammox bacteria), v) aerobic with acetate (optimal for heterotrophic bacteria) and vi) anoxic with acetate (optimal for heterotrophic denitrifying bacteria). Ibuopren was the most biodegradable compound, being significantly degraded under all conditions tested except heterotrophic denitrification. Sulfamethoxazole and metoprolol showed good percentages of removal under certain conditions (up to 70% and 62%, respectively), suggesting the specificity of different microbial groups towards the degradation of these compounds. Finally, carbamazepine and venlafaxine were hardly removed (< 10% in the majority of cases). Results demonstrate that the activation of different microbial groups in combination with altering operational parameters can actually enhance the removal of some of the studied micropollutants.

MO290
Removal of pharmaceuticals in a biofilm reactor: effects of manipulating co-degradation by carbon feeding on system performance L. Zhang, Aarhus University / Department of Bioscience; P. Carvalho, U.E. Bollmann, H. El-taliawy, Aarhus University / Department of Environmental Science; B. Hrix, Aarhus University / Department of Bioscience; K. Bester, Aarhus University / Department of Environmental Science Pharmaceutical contaminants are frequent contaminants in the effluent of municipal wastewater treatment plants as conventional activated sludge systems are unable to completely remove these compounds. Biofilm reactors are a promising biotechnology to remove pharmaceuticals from treated wastewater. Presently, the sand biofilter was operated at 12 h of hydraulic retention time (HRT). In order to prevent adaption of the species composition of the biofilm to the presence of acetate, the system was intermittently fed with influent without carbon addition or with carbon addition. Ten acetate concentration levels were tested in this study, 5, 10, 20, 30, 60, 90, 120, 150, 200 and 300 mg C/L. For each feeding condition (without or with the different carbon concentration), the system was continuously...
operated for 1.5 HRTs, after which four samples were taken over 6 hours. The results showed that with the different acetate additions, the effluent oxygen concentration decreased, reaching the lowest value of 0.98 mg/L at 300 mg C/L acetate addition. However, the oxygen levels in the effluent increased always to initial conditions (4.7 mg/L) in each starving phase between the feeding phases with acetate. The acetate addition resulted in three different compound dependent removal patterns of the micropollutants. Briefly, naphthalene and isothoxazole removal was attributed to co-metabolism (enhanced acetate). Metoprolol, isomerone, diclofenac, propranolol and sulfamethoxazole removal were reduced 1) at lower acetate concentrations by co-metabolic degradation dependent on aerobic turnover, and 2) at higher acetate concentrations limited by suboxic conditions. Moreover, sulfadiazine, sulfamethoxazole and trimethoprim were removed independently of oxygen limitation, which could be interpreted as catabolism. Biofilm reactors can be employed for polishing treated wastewater, and the addition of primary carbon source can enhance the biofilm’s performance.

MO291
Investigating inhibitory effect of anti-inflammatory pharmaceuticals on aerobic sludge metabolism.
M. Green, E. Topuz, G. Yuksel, E. UBay Cokgor, D. Okutman-Tas, Istanbul Technical University / Environmental Engineering
The consumption of pharmaceuticals increase annually due to a variety of reasons involving affordability, population ageing and population growth. Due to the obstacles in monitoring all these micropollutants regularly, focusing on the maximum and daily discharge rate is necessary. To assess the effect of pharmaceuticals on sludge metabolism, there is still need comprehensive research to understand the toxicity mechanism of pharmaceuticals as well as to advance the modelling approaches. A laboratory-scale fill&draw 10L aerobic reactor (sludge age of 5 days; @22°C) was initiated with sludge sample taken from a municipal wastewater treatment plant located in Istanbul. The culture was fed daily with a synthetic wastewater (ISO 8192) (600 mg COD/L) and a synthetic wastewater (ISO 8192) (600 mg COD/L) and acetate (40 mg COD/L). To assess the effect of pharmaceuticals, micropollutants respirometric assays were conducted with pharmaceutical mixture (PMx) as dissolved in MeOH (10, 50, 75g/L of each; Naproxen, Diclofenac, Ketoprofen, Mefenamic Acid, Ibuprofen, Indomethacin). Modelling studies were performed using modified Activated Sludge Model No.1 and AQUASIM2.0 software.

Pharmaceuticals were quantified with LC-MS/MS. Culture amendment with 10μg/L PMx did not result in considerable change compared to control culture, but upon addition of a higher concentration (i.e., 75g/L) there was an increase in the initial substrate consumption rate and decrease in the OUR curve. A removal efficiency in the range of 33-55% was observed for tested pharmaceuticals at the end of the respirometric assays. Measurements showed that pharmaceuticals were not accumulated in sludge phase (+0.2%). Modelling studies reflected that maximum hydrolysis rate of slowly hydrolysable COD (k1) decreased from 0.84 to 0.72 1/day when the concentration of pharmaceuticals increased from 10 to 50g/L. When the concentration of PMx increased from 10 to 50g/L, an increase in the initial COD value of slowly hydrolysable COD was noticed. Furthermore, 75g/L PMx resulted in differentiation in organic matter structure which caused a change in the metabolic rate and hydrolysis rate (k1) can be thus considered as a catalyst. Biofilm reactors can be employed for polishing treated wastewater, and the addition of primary carbon source can enhance the biofilm’s performance.

MO293
Fate and transformation of persistent priority contaminants during potable water reuse: the challenge of producing safe water
C. Raimundo, UNICAMP / Institute of Chemistry; K.H. Cochran, B. Fryer, University of South Carolina; S. Kimura-Hara, University of Calgary; W. Abdelraheem, Y. Huang, University of Cincinnati; S.L. Coffin, University of California, Riverside / Environmental Toxicology; D. Schlenk, University of California, Riverside / Environmental Toxicology; D. Schlenk, University of California, Riverside / Environmental Toxicology; D. Schlenk, University of California, Riverside / Environmental Toxicology; D. Schlenk, University of California, Riverside / Environmental Toxicology; D. Schlenk, University of California, Riverside / Environmental Toxicology.
Elimination of tramadol and methadone in model ozonation experiments: supported by TUBA data for the discharge of these chemicals into the environment. This work is partially supported by SETAC Europe 28th Annual Meeting Abstract Book
MO295
Evaluation of a nano-adsorbent for the removal of metallic carcinogens from wastewater
C. Van der Horst, University of The Western Cape / SensorLab Department of Chemistry; B. Bilwana, Durban university; M. Makonde, Cape Peninsula University of Technology / Chemistry; E. Iwohua, University of The Western Cape / SensorLab Department of Chemistry; V.S. Somerset, CPIT / Chemistry South Africa is experiencing the worst drought in recent history, with water becoming a scarce commodity. However, the supply of good quality water is becoming increasingsely unaffordable. The view of large-scale indirect potable reuse of wastewater is gaining traction in agricultural, domestic and industrial activities. Many technologies including coagulation, membrane process, dialysis, foam flotation, osmosis, photocatalytic degradation and biological methods have been employed for the removal of toxic pollutants from water and wastewater. These technologies are effective but have some disadvantages such as expensive equipment, high operational and maintenance, high energy requirements, generation of toxic residual metal sludge and incomplete metal removal. On the other hand, adsorption offers high efficiency, cost-effectiveness, easy handling and recovery of metals and other adsorbed species. Heavy metals are often found in wastewaters and the removal of these inorganic pollutants using bimetallic iron-based nanoparticles is still unclear. In this study, bimetallic iron-silver nanoparticles were chemically synthesized and immobilized with chitosan to form bimetallic iron-silver nanoparticles (CSFe-AgNPs) to remove heavy metals from wastewaters. In this study, chitosan iron-silver nanoparticles beads have been successfully prepared and its efficiency in the removal of Cd(II) under ambient temperatures has been evaluated. The removal
rate of total Cd(II) from actual wastewater was 89.25%. Furthermore, the monolayer adsorption capacity of Cd(II) based on the Langmuir model was measured to be 90 mg/g. Results were satisfactory when employing the adsorbent for removal of Cd(II) from wastewater samples. Keywords: Adsorption, Bioavailability, Monitoring, Wastewater.

MO296 WATER JPI Project FRAME: A novel framework to assess and manage contaminants of emerging concern in indirect potable reuse

Indirect Potable Reuse (IPR) provides options to maintain sufficient water quotas for a growing population—efficient and affordable strategies for IPR are needed to minimize impacts from a broad range of contaminants of environmental concern (CECs) and to preserve ecosystem services and human health. The project FRAME (A novel framework to assess and manage contaminants of emerging concern in indirect potable reuse) is funded by the European research initiative “Water JPI (Joint Programming Initiative, Water Changing Lives) for a resilient Europe”. The objectives of FRAME are: i) to develop new treatment processes with combined analytical, toxicological and microbiological approaches; ii) to evaluate advanced treatment options in a multiple barrier approach to remove of CECs and inactivation of pathogens; iii) to integrate the experimental results in treatment process models and groundwater models to describe the fate of CECs; iii) to provide a decision support tool for stakeholders, considering process performance and feasibility assessment for treatment scenarios. Advanced treatment options are applied in a multiple-barrier approach at laboratory and full-scale, specifically to improve the removal of CECs, inactivation of pathogens and improvement of other health-related parameters. Detailed fate studies are included to elucidate transformation pathways of CECs and to identify transformation products (TPs) formed during biological processes. Multi-residue analysis methods were developed for sensitive analysis of 176 CECs, including 12 PFAS. The majority of quantitation limits are in the range of 0.5 ng/L to 50 ng/L. Sorption of charged CECs onto Fe-oxides and other minerals was simulated with the goal to create a sorption model for complex soil compositions. Treatment process models using kinetic modelling of CEC and pathogen fate and transport were implemented—designed for integration into a decision support system for stakeholders. Results at laboratory and full-scale showed that a sequential biofilter approach at pilot scale shows higher efficiency than conventional single-stage biofilters; the monitoring of full-scale secondary effluent infiltration sites reveals attenuation of certain CECs, while others require further treatment, highlighting the need for a multi-barrier approach to IPR.

MO297 Evaluation of Rainwater collected from Concrete underground tank and other storage tanks in Owerri Imo State, Nigeria
A. Okeke, University / Chemistry

ABSTRACT Due to the lack of public pipe borne water supply in Owerri municipality and its Environs in Imo State, South-Eastern Nigeria, many individuals have developed different techniques for rainwater storage-harvesting for drinking and water related. Consequently, it is very important to evaluate the quality of rainwater harvested and stored in these different storage tanks so as to ascertain their impacts on rainwater quality. In this study, samples of harvested rainwater were collected from four different storage facilities commonly used by general populace in Owerri (Metal drum tank, concrete underground tank, PVC tank and coated basin for rainwater). The physiochemical and microbiological analysis of these rainwater samples were carried out using standard method. The trace metals in the water samples were relatively below the maximum permissible limit by WHO standard except for lead which was present at low concentration with the value of 0.60 mg/L. The iron content was 0.50 mg/L. For bacteriological analysis, the concrete underground tank recorded the presence of *Pseudomonas* which exceeds the WHO standard stipulated for portable water. The results further explained that concrete underground tank and metal drum tank were more contaminated in terms of physiochemical and microbiological compositions. However, the study shows that harvested rainwater may not be suitable for direct drinking without treatment, but could be used for domestic purposes. Keynote: Harvested Rainwater, microbiological analysis, physiochemical analysis, storage facilities, trace metals

MO298 Sewage Epidemiology: Investigating the Impact of Phthalates on Human Health
C. Allen, L. Jones, Dublin City University / School of Biotechnology and DCU Water Institute; F. Regan, Dublin City University / Chemical Sciences; R.U. Halden, Arizona State University / Biodesign Center for Environmental Security; A. Staines, Dublin City University / School of Nursing and Human Sciences and DCU Water Institute; J. Lawler, Dublin City University / School of Biotechnology and DCU Water Institute

Phthalates are synthetic organic chemicals commonly used as plasticisers in polyvinylchloride and as additives in personal care products. Over 213M kg of phthalates are produced globally each year with end use products including food packaging, paints, tubing and medical devices. Due to their high production volume and continuous release, phthalates are emerging contaminants ubiquitous in the environment. Research has shown that the widespread exposure to these chemicals has been associated with numerous adverse health effects including impaired reproductive health in males, decreased neurological development in children, cancer and obesity. As a result, some phthalates including DBP, BBP, and DEHP have been banned or limited in manufacturing (in particular for items such as children’s toys). As new research emerges indicating that substitute plasticizers also contribute to adverse health effects, these restrictions are likely to increase. The present project constitutes the first application of sewage epidemiology to determine phthalate exposure in an Irish population. Phthalate levels in influent and effluent sewage sludge (biosolids) are being monitored by GC-MS and LCMS/MS analysis, tracking the cycle of phthalates throughout the wastewater system. Phthalate biomarkers are being analysed in influent to assess phthalate exposure. A meta-analysis on health risk data serves to relate the level of exposure to an associated risk, providing the first step in phthalate risk assessment within the Irish environment. Results will inform on the feasibility of using sewage biomarkers for future compliance monitoring. Metabolites from the following phthalates are considered: i) DEHP, dibutylphthalate (DBP), diethylhexylphthalate (DEHP), diisobutylphthalate (DBIP), di-n-octylphthalate (DNOP), diisononylphthalate (DINP), and diisodecylphthalate (DIDP). This study is part of a large-scale project representing an international collaboration between three research centres Dublin City University (DCU), Arizona State University (ASU), and the Norwegian Institute for Water Research (NIVA). With support from Irish utilities, the team is assessing the sources, environmental fates and human exposure profiles of priority phthalates in Ireland. Study results on some eleven priority phthalates will be leveraged to inform risk assessments and environmental policies concerning the phthalate safety and usage.

MO299 Phthalates and their metabolites in the environment
L. Jones, Dublin City University / Biotechnology and Chemical Sciences; C. Allen, Dublin City University / School of Biotechnology and DCU Water Institute; F. Regan, Dublin City University / Chemical Sciences; A. Staines, Dublin City University / School of Nursing and Human Sciences and DCU Water Institute; R.U. Halden, Arizona State University / Center for Environmental Health Engineering; J. Lawler, Dublin City University / School of Biotechnology and DCU Water Institute

Phthalates, or esters of phthalic acid, and their chemical structure consists of one benzene ring and two ester functional groups linked with two consecutive carbons on the ring. These compounds are stable, liquid in ambient temperature, while the ones of higher molar mass are readily insoluble and slightly soluble in water. They are a group of synthetic organic chemicals that are used as additives, or plasticisers, to enhance the flexibility, transparency, stability, longevity, and durability of plastic materials and as non-plasticisers in consumer products. They are most commonly used as plasticisers in polyvinyl chloride (PVC) and phthalates have been used in the plastics industry for more than 80 years. As phthalates are so commonly used, their impact on the environment and human health has been extensively studied. Phthalates have been found to be recalcitrant, ubiquitous within the environment, and in many cases, detrimental to human and animal health. This project represents an important collaboration between three research centres (DCU, ASU, & NIVA) with support from Irish Water, Panda and Ficklin Co., to assess the potential sources and environmental fates of priority phthalates in Ireland. This project is supported by several by studies carried out in Ireland already (DCU) and a vast array of literature in the area of priority pollutant monitoring. The impact of such study would be the analysis of these eleven phthalates from source to fate, in order to inform environmental policy on the risks posed by phthalate usage. The phthalates included in this study are Dibutylphthalate (DBP), Dipropylphthalate (DPP), Diisobutylphthalate (DPBP), Diisopropylphthalate (DIPP), Dimethylphthalate (DMP), Diethylhexylphthalate (DEHP), Di-n-octylphthalate (DNOP), Dioctylphthalate (DOP) and Dimethylphthalate (DMP). A selection of phthalate monoesters have also been included in this study to evaluate human exposure to phthalates in Ireland already (DCU) and a vast array of literature in the area of priority pollutant monitoring.
extent of phthalate contamination within Ireland, and the impacts on human health, are unknown.

**MO300**

Poly- and perfluoroalkyl substances (PFASs) in the sewage system of the Bordeaux city: high contribution of unidentified precursors of perfluoroalkyl acids.

C. Simonnet-Laprade, University of Bordeaux UMR EPOC; P. Labadie, UMR CNRS EPOC Universite Bordeaux / UMR 5805 EPOC; M. Capdeville, LyRE Centre de Recherche et Développement SUEZ; P. Pardon, UMR CNRS EPOC Universite Bordeaux / EPOC UMR 5805; H. Budzinski, University of Bordeaux This study proposes to identify the origin of 30 poly- and perfluoroalkyl substances (PFASs) entering a French city (Bordeaux Metropolis). For this purpose, 16 samples of domestic wastewaters, 10 samples of wastewaters impacted both by industrial and commercial activities were collected within the sewage network upstream typical and representative treatment plants; in addition 4 samples of runoff waters were also targeted in order to explore the input of this type of potential source. PFASs were also analyzed in the influents, the effluents, and the sludges of the 4 main wastewater treatment plants (WWTP) of Bordeaux Metropolis to quantify global inputs to the natural aquatic environment. The results highlight distinct patterns and levels of contamination between different types of samples and potential sources. Overall, wastewaters impacted by industrial inputs have the highest levels (ΣPFAS = 4.6-501.7 ng L⁻¹) with the predominance of PFOS, PFHxS, C₆-C₇ PFCA and 6-2 FTSAs. High levels of 8.2 and 10.2 FTSAs (> 100 ng L⁻¹) were found in wastewaters from the harbor area and a major industrial area. Domestic wastewaters have the highest levels of 6.2 dpIP (median concentration of 4.5 ng L⁻¹), probably related to its use in food packaging. Concerning runoff waters, the highest levels are found in the airport area with ΣPFAS of 227 ng L⁻¹. The comparison of the profiles with those found for WWTP influents using a principal component analysis made it possible to highlight the influence of the industrial and commercial discharges on the global contribution to WWTPs. It has been possible to quantify global fluxes of PFASs to the four WWTPs and they were estimated at about 14 g d⁻¹ for the sum of targeted PFASs; concerning removal in WWTPs, only the C₆-C₇ PFCA, the PFOS and their precursors with more than 8 perfluorinated carbons were shown to be partially or totally removed by the treatment processes. In addition to the targeted analysis and in order to estimate the proportion of unidentified perfluoroalkyl acid precursors, the total oxidizable precursors (TOP) assay developed by Houtz and Sedlack (2012) was applied to each of the matrices. This method, which was applied for the first time to domestic wastewaters and to WWTP sludges, revealed the presence of unidentified precursors of C₆-C₇ PFCA in all the matrices representing 32 % (in runoff water) to more than 90 % (in domestic wastewaters) of the total PFAS molar concentrations.

**MO301**

Antibiotics and endocrine disrupting compounds in wastewater treatment plants and in receiving water bodies around the city of Rome (Italy) 

I. Ruffo, National Research Council / Institute of Water Research IRIS-CNRC; F. Spataro, N. Ademollo, T. Pesca lure, National Research Council of Italy / Water Research Institute; L. Patrolecco, Water Research Institute-National Research Council / Water Research Institute Several studies highlighted the occurrence of organic micropolutants such as pharmaceuticals residues, ingredients of personal care products and endocrine disrupting compounds in wastewaters and receiving surface waters in Italy. In this work, we aimed at analyzing concentrations ranging from ng L⁻¹ to few mg L⁻¹. Among the emerging compounds, antibiotics are of peculiar interests due to their potential adverse effects on aquatic ecosystems diversity and function and because they can act as a potential driver for dissemination of resistance genes. Previous studies show that classes of synthetic antibiotics, such as quinolones, sulfonamides, tetracyclines, betalacitams and macrolides, widely used in human and veterinary medicine, are rather resistant to microbial degradation, providing an indication as to why these compounds might persist within wastewater treatment plant (WWTP). Nonylphenol ethoxylates (NPEOs), commonly used as detergents, wetting agents, emulsifiers and dispersants, are precursor of nonylphenols (NPs) by the loss of the ethoxy groups. These compounds are biocidal because they can bind to estrogen receptors and can block or alter endogenous estrogen functions in various reproductive and developmental stages. In fact, NPs have been included in the European list of priority hazardous substances for surface waters in the Water Framework Directive. The aim of the present study was to investigate the occurrence and fate of selected classes of emerging and ED compounds in four WWTPs serving the city of Rome and in the receiving clean effluent from WWTPs around the city of Rome and in contaminated sites along the urban stretch of Tiber and Aniene rivers. The results confirmed that WWTPs were the main source of river contamination. Although the efficient wastewater input into receiving water should produce a dilution of contamination, the continuous release of these xenobiotics into the aquatic environment may lead to chronic exposure of organisms at all levels of the food chain.

**MO302**

Mass flows of antimicrobial compounds in Swedish sewage treatment plants 

M. Ostman, J. Fick, M. Tysklind, Umea University / Department of Chemistry Antimicrobial biocides are used as disinfectants, antiseptics and preservatives to prevent unwanted microorganisms. In the same manner as antibiotics, they are entering our sewage system and passing on to the sewage treatment plants. Sewage treatment plants has been suggested as a possible high-risk environment when it comes to development of antibiotic resistant bacteria. Concerns has been raised that biocides might promote antibiotic resistance via co- and cross-resistance mechanisms. It is therefore important to quantify and understand the fate of these compounds within the sewage treatment plants to be able to design and construct more efficient future plants, as well as an aid in risk assessment of these chemicals. The aim of this study was therefore to determine detailed mass flows of eleven antimicrobial compounds within three Swedish sewage treatment plants. In total, the three plants were sampled for nine days for samples representing the major flows in respective plant i.e. incoming wastewater, treated effluent, wastewater after the primary clarifier, primary sludge, surplus sludge, digested sludge. All samples were analysed by liquid chromatography tandem mass spectrometry (LC-MS/MS). The compounds included different quaternary ammonium compounds (QACs), such as 12-BAC, 14-BAC, DDMAC , CPC and CTAB as well as other compounds such as chlorhexidine, benzotriazole, ciprofloxacin and fluoroanole. QACs and chlorhexidine were effectively removed in the treatment process, whereas most of the other compounds were recovered after the primary clarifier or to the digested sludge. The major yearly loads in the treated effluent in three studied plants was 29.4 kg and 2900 kg in the digested sludge. For more polar compounds such as trimethoprim and fluoroanole, about half of the amount found in the incoming sewage water was found in the treated effluent. To our knowledge this is the first detailed mass balance study for QACs and chlorhexidine reported. The study provides not only new scientific understanding but also important knowledge to e.g. sewage treatment plant operators and law- and policy makers.

**MO303**

Herbicides and fungicides in wastewaters of agricultural regions of Ontario T. Sultana, Trent University / Environmental and Resource Studies; P.A. Helm, Ontario Ministry of Environment and Climate Change / Environmental Monitoring and Reporting Branch; C.D. Metcalfe, Trent University / Water Quality Centre Herbicides and fungicides are widely used in agriculture to control weeds and fungal diseases that can reduce crop yields. There is potential for these compounds to be transported from treated fields into surface waters via agricultural runoff. The objective of this project was to evaluate the distribution of selected current-use fungicides and herbicides in 5 major rivers and 13 smaller streams within regions of intense agriculture in southern Ontario, Canada. The Polar Organic Chemical Integrative Sampler (POCIS) was selected as a principal monitoring technique, although grab samples of surface waters were also collected throughout the POCIS deployment periods. The sampling rate for each target compound (90% reduction) but the majority remained in the digested sludge. The total yearly loads in the treated effluent in three studied plants was 29.4 kg and 2900 kg in the digested sludge. For more polar compounds such as trimethoprim and fluoroanole, about half of the amount found in the incoming sewage water was found in the treated effluent. To our knowledge this is the first detailed mass balance study for QACs and chlorhexidine reported. The study provides not only new scientific understanding but also important knowledge to e.g. sewage treatment plant operators and law- and policy makers.

**MO304**

Antimicrobial biocides might promote antibiotic resistance via co- and cross-resistance mechanisms. It is therefore important to quantify and understand the fate of these compounds within the sewage treatment plants to be able to design and construct more efficient future plants, as well as an aid in risk assessment of these chemicals. The aim of this study was therefore to determine detailed mass flows of eleven antimicrobial compounds within three Swedish sewage treatment plants. In total, the three plants were sampled for nine days for samples representing the major flows in respective plant i.e. incoming wastewater, treated effluent, wastewater after the primary clarifier, primary sludge, surplus sludge, digested sludge. All samples were analysed by liquid chromatography tandem mass spectrometry (LC-MS/MS). The compounds included different quaternary ammonium compounds (QACs), such as 12-BAC, 14-BAC, DDMAC , CPC and CTAB as well as other compounds such as chlorhexidine, benzotriazole, ciprofloxacin and fluoroanole. QACs and chlorhexidine were effectively removed in the treatment process, whereas most of the other compounds were recovered after the primary clarifier or to the digested sludge. The major yearly loads in the treated effluent in three studied plants was 29.4 kg and 2900 kg in the digested sludge. For more polar compounds such as trimethoprim and fluoroanole, about half of the amount found in the incoming sewage water was found in the treated effluent. To our knowledge this is the first detailed mass balance study for QACs and chlorhexidine reported. The study provides not only new scientific understanding but also important knowledge to e.g. sewage treatment plant operators and law- and policy makers.

**MO305**

A Study on the Distribution and Behavior of Nonylphenol in the Suyeong River, Korea D. Kim, Pukyong National University / Department of Ecological Engineering; S. Cho, National Institute of Fisheries Science; M. K. Ro, Pukyong National University / Department of Environmental Engineering; Y. Kim, Pukyong National University / Department of Food science and Technology; Y. Chung, Pukyong National University / Department of Ecological Engineering Nonylphenol is known, as one of Endocrine Disrupting Chemicals is the degradation product of Nonylphenol ethoxylates being used as nonionic surfactant. Nonylphenol is classified as an endocrine disrupter capable of interfering with the hormonal system of numerous organisms. In order to understand the current contamination and behavioral characteristics of Nonylphenol by measuring the concentration of Nonylphenol in the surface water in the downstream of Suyeong...
River of Korea, and based on that, estimating the material balance. During the survey period, the range of Nonylphenol concentration in estuary of Suyeong River had the range of 142.0 – 569.6 ng/L and the average of 271.0 ng/L. The target area was divided into 3 regions to estimate the material balance of Nonylphenol in the downstream of Suyeong River. The dissolved Nonylphenol of 282.3 g/day occurred in region 1 of Suyeong River, and Nonylphenol influx load occurred in particular suspended solids of 125.0 g/day. The dissolved Nonylphenol outflow discharge toward the region 2 was 192.5 g/day, while the adsorption to the particulate suspended solids was 89.8 g/day. In the case of Nonylphenol within the particulate suspended solids, the outflow to the region 2 was 1,250.0 g/day and the amount of settling toward the sediment was estimated to be 422.7 g/day. The adsorption from the dissolved Nonylphenol to the particulate suspended solids in the region 1 and region 2 was 31.8% and 54.9%, respectively. In the region 3, the desorption rate was 8.8%.

MO306 Drugs of abuse distribution in Turia River based on geographic information and ecotoxological assessment
M. Andrés Costa, University of Valencia / Environmental and Food Safety Research Group; J. Pascual-Agullar, V. Andreu, CIDE CSIC UV GV; Y. Pico, University of Valencia / Medicine Preventive
The aim of this study establishes the influence and the effects of the human pressure in a typical Mediterranean River Basin to determine accurately the point sources of contamination through an environmental forensics methodology. A total of 42 drugs of abuse and a number of the six parameters with the highest level of data quality were determined from 22 sampling sites in 2012 and 31 in 2013 distributed along the river. Analysis of the target illicit drugs was performed using solid phase extraction and liquid chromatography coupled mass spectrometry (SPE-LC-MS/MS). To determine spatial incidence of drugs of abuse, analytical results of target compounds were georeferenced and integrated into a geographical information systems (GIS). Ecological risk assessment of drugs of abuse distribution was performed using the model MePeTox2000, calculated by applying risk quotient (RQ). In 2012, 3,4-methylenedioxyamphetamine (MDMA) and 4-methoxycyclphenidine (4-MeO-PCP) were detected in one sampling point at a concentration of 22.8 and 37.6 ng/L, respectively. In 2013, 4-MeO-PCP was detected in a different sampling point of 2012 at a concentration of 7.55 ng/L and cocaine methyl ester (ECME) was detected at a concentration of 15.03 ng/L. Butofenate (BUF), methadone (MET) and p-methoxymetamphetamine (PMA) were found out in 3 or 4 sampling points at concentrations < 10 ng/L in 2012. Ephedrine (EPH) and codeine (COE) were detected in 3 sampling points at average concentrations of 11.6 ng/L for EPH and 91.3 ng/L for COD in 2013. The compound detected more frequently along the river was benzoylecgonine (BECC), a cocaine metabolite, with an average concentration of 25.4 (2.91–76.8) ng/L in 2012. In 2013, MDMA was detected in 5 sampling points (mean of 4.67 ng/L ranging from 2.34 to 7.21 ng/L and BECG and MET were detected in a total of 8 and 7 sampling points, respectively, each one at a mean concentration of 14.02 (1.83–12.7) ng/L for BECG and 11.4 (2.29–40.1) ng/L for MET. GIS provided the spatial incidence of drugs of abuse along the Turia River Basin. The occurrence of drugs of abuse could be correlated with water quality indexes and a higher number of bars and restaurants located close to the Turia River Basin. 

MO307 Occurrence, fate and environmental risk assessment of benzenophene-type UV filters in a tropical urban watershed
K. Gin, National University of Singapore / Civil & Environmental Engineering; F. Mao, National University of Singapore / Civil and Environmental Engineering; L. You, National University of Singapore; M. Reinhard, Stanford University; Y. He, Shanghai Jiao Tong University
A variety of benzophene compounds (BPs) have been used as ultraviolet (UV) light absorbers in personal care products and synthetic products that are exposed to sunlight. Following use, BPs can enter ambient environments directly via treated or untreated wastewater discharges. Numerous studies have been conducted to detect the occurrence of BPs in the aquatic environments. However, the occurrence and fate of BPs in tropical waters remains poorly understood. In this study, occurrence and fate of seven BP-type UV filters (i.e., 2,4-dihydroxybenzophenone (BP-1), 2,2',4,4'-tetrahydroxybenzophenone (BP-2), 2-hydroxy-4-methoxybenzophenone (BP-3), 2',2'-dihydroxy-4,4'-dimethylbenzophenone (BP-6), 2',2'-dihydroxy-4-methoxybenzophenone (BP-8), 4-hydroxybenzophenone (4OH-BP) and 4,4'-dimethoxybenzophenone (4DBH)) were investigated in a tropical urban watershed consisting of five major tributaries that discharge into a well-managed water body. The BPs concentrations were measured in four compartments, i.e., bulk water, suspended solids, pore water and sediments. Results showed that benzophene concentrations varied from widely < LOQ to 122.6 ng L\(^{-1}\) in dissolved phase and < LOQ to 2774 ng L\(^{-1}\) in solid phases. Suspended solids in the water column contained significantly higher amounts of BPs than sediments, while the concentration difference between bulk water and pore water was insignificant. Further study will evaluate the vertical concentration profile in the aqueous phases and in the solid phases. The concentration ratio of BP-1 to BP-3 will also be addressed, aiming at the evaluation of degradation of BP-3 in the field. This will be followed by a preliminary risk assessment.

MO309 Formation of disinfection byproducts throughout various drinking water treatment processes
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This work investigates the formation of disinfection byproducts (DBPs) throughout the treatment processes operating in the various drinking water treatment and desalination plants and distribution system that supply drinking water to more than 4.5 M inhabitants living in the city of Barcelona and its metropolitan area. For this, DBP formation potential tests with chlorine were performed with the water entering each plant and produced after the individual processes carried out in each plant to treat the water. DBP mixtures were generated for each investigated matrix at two different temperatures (15°C and 25°C) and reaction times (0, 24 h, and 48 h, and also 72 h in the plant effluent) so that different scenarios in the drinking water distribution network were simulated. Six different DBP classes in total were investigated in the DBP mixtures generated by means of gas chromatography coupled to mass spectrometry detection. The list included the regulated N-nitrosodimethylamine (NDMA) and the regulated N-nitrosamines formed during chloramination. Moreover, NDMA can also form through ozonization or nitrosation. Nitrosamines can form in water in specific conditions. N-nitrosodimethylamine (NDMA) is a potential carcinogen and is found in trace amounts in chlorinated drinking water. NDMA is the most widely detected N-nitrosamine in drinking water which was subjected to chloramination. NDMA is also found in water subjected to disinfection with chlorine, hypochlorite or other chlorine-generating compounds, but in lower amounts. NDMA can also form through ozonation or nitrosation. Multiple mechanisms can be involved in NDMA formation: a) reaction between inorganic nitrite or dinitrite and aromatic amines; b) reaction with typical secondary amine precursors; c) chlorination of nitrite in the presence of nitrosamine precursors; d) catalytic formation on activated carbon, from secondary amines; e) UV or sunlight photolysis of nitrite, in the presence of secondary amines. Many organic nitrogenous substances can be NDMA precursors (pharmaceuticals, substances used in cosmetics, pesticides, chelating agents, amine-based polymers, etc.), but not all can be present in significant amounts in the source water. In the context of water treatment for potable use, amine-containing coagulation polymers and some anion exchange resins constitute the main source of NDMA precursors. Although NDMA is the most prevalent nitrosamine detected, according to the surveys conducted until now, it may account for only a minor fraction of all nitrosamines formed during chloramination. More research is required in order to establish ways to avoid NDMA and other nitrosamines formation.

MO310 Formation of N-nitrosodimethylamine during water treatment for potable use: an update
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Nitrosamines can form in water in specific conditions. N-nitrosodimethylamine (NDMA) is a potential carcinogen and is found in trace amounts in chlorinated drinking water. NDMA is the most widely detected N-nitrosamine in drinking water which was subjected to chloramination. NDMA is also found in water subjected to disinfection with chlorine, hypochlorite or other chlorine-generating compounds, but in lower amounts. NDMA can also form through ozonation or nitrosation. Multiple mechanisms can be involved in NDMA formation: a) reaction between inorganic nitrite or dinitrite and aromatic amines; b) reaction with typical secondary amine precursors; c) chlorination of nitrite in the presence of nitrosamine precursors; d) catalytic formation on activated carbon, from secondary amines; e) UV or sunlight photolysis of nitrite, in the presence of secondary amines.
Toxicology and Risk Assessment TAYER / Rey Juan Carlos University Historically, coastal and transition waters have preferentially been used for human settlement, leading to a continuous input of wastewater effluents into these habitats. Water quality monitoring of these systems, with the goal of improving their protection under the water framework directive is highly needed. The number of studies characterizing the presence of psychoactive pharmaceuticals (PAs) in coastal waters and marine biota is still limited. Our work represents the first attempt to study these compounds in the Rias Baixas area (North Western Spain). This area was chosen as the location for this study due to its economic and ecological importance as a national and global leader in shellfish (especially mussels) production. In this study, the presence of 16 PAs (benzodiazepines and anxiolytics) was studied in samples of sea water and tissue from 7 economically valuable and highly consumed marine species. Samples were collected from 22 sites along the three main Rias Baixas. Fourteen out of the 16 assessed substances were detected in the water samples with venlafaxine (64%) and citalopram (41%) showing the highest detection frequencies. The highest concentrations in water were also measured for venlafaxine (291 ng/L), followed by lorazepam (95.90 ng/L) and citalopram (92.50 ng/L). Only 3 PAs (alprazolam, citalopram and venlafaxine) were present in the collected biota samples (razor clam, clam and octopus) in concentrations up to 14 ng/g d.w. (citalopram), 3 ng/g d.w. (venlafaxine) and 0.31 ng/g d.w. (alprazolam). Hazard quotients (HQ), calculated from the measured concentrations in water and available chronic aquatic toxicity data resulted in values higher than 1 (indicating elevated hazard and possible risk) for venlafaxine, citalopram, and sertraline. The venlafaxine concentration) by glyphosate and AMPA. Fish were fished in the Marne River (a tributary of the Seine River) by electrofishing. Although it is not bioaccumulative, the associated use of surfactants in pesticide formulations allows better assimilation in biological tissues. The aim of our study was first to develop a method to assess glyphosate and AMPA levels in biological tissues and then to determine the contamination of a freshwater fish, the European chub (Squalius cephalus) by glyphosate and AMPA. Fishes were fished in the Marne River (a tributary of the Seine River) by electrofishing. Although it is not bioaccumulative, the associated use of surfactants in pesticide formulations allows better assimilation in biological tissues. The aim of our study was first to develop a method to assess glyphosate and AMPA levels in biological tissues and then to determine the contamination of a freshwater fish, the European chub (Squalius cephalus) by glyphosate and AMPA. Fishes were fished in the Marne River (a tributary of the Seine River) by electrofishing.

New Horizons in Particulate Polymer Analysis: Micro- and Nanoplastics and Tire Rubber Detection, Characterisation and Impacts in the Environment (P)

MO315 MPHunter: a dedicated software for µFTIR- Imaging Microplastic data analysis. First development steps and future perspectives A. Vianello, Aalborg University / Civil Engineering Department - Section of Water and Environment; S. Primpke, G. Gerds, Alfred Wegener Institute / Shelf Sea System Ecology; M. Simon, N. van Alst, K.B. Olesen, F. Liu, J. Vollertsen, A. Vianello, Aalborg University / Civil Engineering Department Imaging-Micro-Fourier Transform Infrared Spectroscopy (Imaging-µFTIR) applying a Focal Plane Array (FPA) is the most promising analytical tool for direct and rapid analysis of microplastics (MP) deposited on IR windows or collected on suitable membrane filters. The amount of data produced during an imaging analysis (many GB), combined with the lack of specific features addressed to MP identification and quantification in commercial FTIR analysers, makes the analysis of the IR map extremely time-consuming and partially operator dependent. Although a novel automatic analysis pipeline has already been developed by Primpke et al. (2017), the spectral identification is still performed using a commercial FTIR software, limiting the use of the pipeline to the FTIR software's owners. Here we present a dedicated software (MPHunter) for MP analysis which can export, convert and manage datasets from the two FPA-µFTIR Imaging suppliers. The software, which can manage several million single spectra and many reusing an unwanted resource and the economic advantage for farmers who have to pay little or nothing to use the resource. Furthermore it has been shown that wastewater pollutant load can be reduced as it goes through the environment through processes such as photolysis, biodegradation and adsorption. Using these natural processes to our advantage can reduce the costs of treating wastewater. However it has been shown that treated and untreated wastewater contain emerging pollutants (e.g. pharmaceuticals, personal care products, antibiotics, hormones, etc). When reusing wastewater for irrigation we are creating a pathway for these pollutants to enter the environment and possibly the human and animal food chains. Therefore to adequately assess this practice it is necessary to have a clear understanding of the presence, fate and prevalence of emerging pollutants from source (irrigation water), through soil and finally in plant tissue. Therefore this paper presents an analytical relevant environmental matrices (i.e. water, soil and plants). This method will help determine the risk posed to humans, animals and the environment when reusing wastewater for irrigation. Furthermore it will aid in evaluating the natural attainment of emerging pollutants in the agricultural environment and therefore inform on the level of treatment necessary to undertake this practice sustainably.
thousands of reference spectra in one run, is written in RAD Studio (Embarcadero Delphi IDE), an object-oriented programming environment which allows easy construction of user-friendly interfaces. As an example, an imaging dataset of 196 tiles from a 128x128 pixel FPA detector (totaling 3.2 million individual spectra) can be easily managed using the software’s features. The software calculates the Pearson’s correlation coefficient between the unknown spectra and a reference spectral library. The correlation coefficient can be further refined to define particles. The reference spectra can be easily created and uploaded to the software as a .csv file. The calculation time for comparing 3.2 million spectra to a library of 150 spectra is around 6 hours on a standard laptop. Software features include conversion from %Transmittance to Absorbance and vice versa, selection of multiple customizable spectral ranges/whole spectral range for correlation and filters for residual noise removal. The correlation results can be further refined to define particles. Potential MP can then be marked, measured (main axes, area) and saved. MPHunter is open source freeware. It allows a semi-automated MP identification and quantification, decreasing the time demand for the interpretation of FTIR-imaging data and increasing the data accuracy. Further improvements are ongoing to interface MPHunter to the Analysis Pipeline from Primpke et al. (2017) and use it as the searching engine.

MO316
From alpine regions to dense populated areas: A comparison of microplastic contamination between 15 rivers across Germany
Among marine litter, plastic waste is of growing concern, as nowadays it has become ubiquitous in the oceans. A large portion of the plastic waste is produced onshore and then enters the marine environment via water discharge to the river systems. Although, the oceans are considered as the main sink of plastic debris, recent studies also reported on the contamination of freshwater ecosystems with microplastics. Therefore, freshwater ecosystems do not only act as a source of plastic particles for the oceans, they also act, at least temporarily, as a sink. This may come along with all the associated harmful consequences that have been reported previously for marine ecosystems. Nevertheless, there is a considerable gap of knowledge about the impact and contamination of freshwater ecosystems with plastic particles. The lack of harmonized methods for microplastics sampling and detection hamper the comparability of data on concentrations and the composition of synthetic polymers in the freshwater environment. We compared microplastic contamination down to 20µm between 15 rivers across Germany, by the use of a harmonized sampling, sample processing and sample analysis (FTIR) currently developed by the IFU (projects: FRES and BASIEAN). Results show a high variability between rivers and due to the lower size fraction measured relatively high concentrations compared to the few studies conducted in surface waters of freshwater ecosystems so far. Further, our data may shed light on major pathways and sources of microplastics in freshwater ecosystems. (M. Loeder, I. Schrank and H. Imhof contributed equally to the work as first co-authors).

MO317
Analytical approach for the identification and quantification of microplastic particles in environment samples by particle analysis in combination with FTIR and Raman microscopy
The detection of microplastic particles in an environment sample in the wide range from 1 µm to 5 mm nearly quantitatively in a reasonable time is a challenging mission. This task has to be handled with well-defined sampling procedures and sampling locations. Next task is the sample preparation procedure to remove organic and inorganic parts in such a manner, that the microplastic particles will not be destroyed. A further important point is the subsequent loss free transport to an analytical lab and the use of blind samples from the sampling location through all steps until the lab. Starting from this point our poster describes the following pratical steps to identify microplastic particles by the methods used in environment samples. All operations and analyses are performed in dust-free rooms in flow boxes and all equipment is plastic-free. First procedure is a three-step vacuum filtration to divide the particles in four fractions, above 500 µm, from 500 to 50 µm, from 50 to 10 µm and below 10 µm. After filtration the particles of the different fractions are on silicon filters made from wafers which are IR transparent. Now optical particle identification programs determine all particles in shape and dimension and store there coordinates for the subsequent measurements with FTIR and Raman microscopy. Both methods identify the microplastic particles on the filter by their chemical structure using spectral databases. This spectral search runs automatically. The databases include polymers, copolymers, fillers, inorganic and organic substances, paints and lacquers. The IR libraries use transmission, ATR and reflection spectra and the Raman libraries use spectra measured with 532, 633 and 785 nm lasers. Every particle is assigned about the spectrum to a substance. This can be a polymer or in case of a mixed spectrum a polymer with paint/filler or also not a polymer. The fractions above 500 µm and from 500 to 50 µm are measured manually with FTIR and the fractions from 50 to 10 µm and some selected samples below 10 µm mainly by Raman. However, several samples of the fractions above 10 µm are measured also with both methods since a combination of both delivers complete microplastic particles analysis. Further parameters, as the integration time, the number of accumulations, the magnification of the objectives and the lateral resolution, which influence the results concerning measurement time and locating and identification of mainly smaller particles will be discussed.

MO318
Using pyrolysis GC-MS in combination with multivariate tools to identify and differentiate polymer type and weathering of microplastics
T. Storslet, L. Sorensen, K. Almaas, SINTEF Ocean / Environmental Technology; M.O. Høysæ, Norwegian University of Science and Technology; O. Brasket, A. Booth, SINTEF Ocean / Environmental Technology.
Pyrolysis gas chromatography coupled to mass spectrometry (pyGC-MS) is a promising tool for identifying and quantifying trace amounts of microplastic (MP) in environmental samples. For pristine plastic samples, it has been demonstrated that polymer type and additive chemicals can be elucidated from the obtained pyrograms and their underlying mass spectra. In this study, an automated method for MP classification was developed. Pyrograms obtained from environmental samples are typically complicated by the presence of naturally occurring organic compounds and the presence of multiple polymer types. Furthermore, weathering processes such as oxidation and biodegradation may alter the chemical composition of the polymers, especially at the surface. Thus, an updated analysis approach was first used to classify pristine and environmental MP samples. Multivariate tools were then applied to classify the samples based on the global pyGC-MS derived composition of the polymers, and to compare pristine materials with samples from the environment. The technique shows promise where manual techniques fail or have difficulty due to the lack of visual resolution of chromatographic peaks with important diagnostic mass spectral features.

MO319
Marine Microplastic: Production and characterisation of realistic test materials for studying ecosystem impacts
Reported studies investigating the possible effects of plastic litter on marine biota have almost exclusively utilised pristine plastic materials that are homogenous in polymer type, shape, size and composition. The production of realistic test materials for microplastics (marine litter < 5mm), as collecting samples of such material from the marine environment in quantities sufficient for use in laboratory impacts studies is simply not feasible. Crucially, weathered plastics collected from the marine environment show considerable physical and chemical differences to pristine and post-production consumer plastics. In the current study, we describe the preparation and characterisation of a more environmentally realistic marine litter-derived microplastic reference material (≤5 mm) for use in fate and effects studies. Weathered marine plastic litter (351 items) was collected from the coast of the island of Texel (The Netherlands) and carefully identified and categorised (fibre-based, packaging, foam, plastic boxes and jerry cans, bottles, gloves and miscellaneous plastic materials). From the collected material, comprising ~70% of the collected material, which contained 9 different polymer types. The macroplastic material was sub-sampled and subjected to a cryo-milling and sieving process to create the microplastic reference material. To confirm that the original macroplastic polymer distribution was mirrored in the generated microplastic sample, it was subjected to ATR-FTIR and differential scanning calorimetry analysis. Particle size distribution (PSD) of the microplastics from the pristine and weathered plastics showed that 68% (by mass) of the particles were in the range between 0.5 and 2.0 mm. Particle number increased with decreasing particle size fraction. Scanning electron microscopy revealed a wide range of particle sizes and shapes reflecting the properties of the different polymers. ICMP-MS and ICP-OES analyses revealed the presence of a broad range of metals and other elements (e.g. Al, Cr, Fe, Mg, Pb, S and Zn) associated with the final sample. Many of these represent common inorganic plastic additives used as colourants, fillers and stabilisers. The additive organic chemical profile of the macroplastic mixture was also determined by GC-MS analysis following extraction by ethyl acetate and ultrasonication. A broad
MO320 Optimization of the preparation of standards of high density polyethylene microplastics and quantification techniques by stereoscopic and confocal microscopy.

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A non-complex procedure has been developed for preparing HDPE microplastics as standard for microplastic determination in sediments. Always keeping ensuring minimal contamination in mind, small pellets and bottle caps from several brands were studied in order to identify those that could provide a clear spectrum for HDPE using Raman spectroscopy (considering that Raman spectroscopy is sensitive to those additive and pigment chemicals in microplastics that interfere with the identification of polymer types). Red caps from a popular brand of mineral water were selected as the raw material as their spectrum was easily comparable with those provided in the literature for HDPE. The large pieces of plastics were converted into microplastics by using a conventional machining process, i.e., a drill with a sandpaper implement (Dremel 300, 13 mm-60 grain size sandpaper). With this approach, several sequential sessions of the machining processes were carried out in order to avoid changes in the physical properties of the plastics that would result from temperature increase due to due to prolonged friction. After the machining, the obtained HDPE particles were sieved using ethanol (96%) through two different mesh sizes until a final standard ranging from 0.1 to 0.850 mm was achieved. This range was selected as it is relevant in biota ingestion risk as well as is appropriate for visual counting using microscopes. The standard HDPE microplastics used in all the extraction experiments were the same. It was thoroughly mixed previously to spiking in order to avoid differences in the distribution of sizes. To prepare a standard for HDPE microplastics were suspended in ethanol 96% and shook using a magnetic stirrer. An effective method for determining the particle distribution of microplastics is microscopy. In this work, two types of quantification using microscopy were used and compared: optical microscopy by visual sorting (Leica ICC50 HD, 4X lens, using a mesh for counting with sections of 3x3 mm prepared for this study) and, confocal microscopy (Zeta Instruments, model Zeta 300). The last one, included object detection algorithms (Mathematica 10) which not only allows quantification of plastic particles but also their classification into size groups.

MO321 First Report of Microplastics in Pacific-side Arctic Ocean

H. Lee, S. Kim, Incheon National University / Department of Marine Science; S. Kang, Korea Polar Research Institute / Division of Polar Ocean Science

The Arctic is one of the pristine areas that are sensitive to global environmental changes and have a relatively low environmental pollution. However, Arctic is already affected by floating microplastics (MPs) according to previous studies (38-234 pieces of sea ice and 8.34 pieces/cm² in the Arctic polar water). Previous research on the Arctic has concentrated on the waters associated with the Atlantic Ocean (for example, the Barent Sea), while the Arctic Sea (e.g., the Chukchi sea, East Siberian sea, etc.) linked to the Bering strait has never been studied. To prepare a standard, HDPE microplastics were suspended in ethanol 96% and shook using a magnetic stirrer. An effective method for determining the particle distribution of microplastics is microscopy. In this work, two types of quantification using microscopy were used and compared: optical microscopy by visual sorting (Leica ICC50 HD, 4X lens, using a mesh for counting with sections of 3x3 mm prepared for this study) and, confocal microscopy (Zeta Instruments, model Zeta 300). The last one, included object detection algorithms (Mathematica 10) which not only allows quantification of plastic particles but also their classification into size groups.

MO322 Biodegradability of pristine and weathered car tire rubber using different inocula

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Car tire wear is estimated to represent a major fraction of microplastic pollution in the environment. Rubber particles are transported by wind transport and road runoff, thereby reaching soils and wastewater treatment systems. Information on their occurrence, fate and degradability in environmental and engineered systems is limited yet crucial to determine their persistence and potential risks associated to their exposure. In this study, we assessed the biodegradability of car tire rubber (90-125 µm) under controlled laboratory-scale conditions. Standardized batch tests (OECD 301 and ISO 14851) were used to determine complete mineralization under
aerobic conditions of pristine and UV-weathered tire rubber in the presence of three different microbial inocula, i.e. activated sludge, soil particles and soil supernatant. Acetate and poly(D,L-lactide-co-glycolide) (PLGA) were used as positive controls in terms of readily degradable substrate and degradable plastic material, respectively. Pristine and weathered rubber exhibited low but measurable biodegradation levels in the presence of activated sludge (3.8–7.6% THOD) and soil supernatant, while no significant biodegradation was observed when using soil particles as inoculum. Accelerated UV weathering and higher biomass concentration was found to increase the biodegradability by activated sludge. Interestingly, increased response in terms of oxygen consumption was observed with acetate as co-substrate, indicating improved rubber degradability in the presence of a readily degradable carbon source. PLGA exhibited limited biodegradability and the addition of electron donors to the system increased porosity and roughness on rubber surfaces over the course of the experiments, seemingly indicating degradation via surface colonization. Overall, ready biodegradability tests proved suitable to obtain information on degradation of car tire rubber. This study provides first evidence of their degradability, especially for weathered rubber in the presence of a co-substrate, which should be considered for future detoxification assessment. Noteworthy, exposure of rubber to microbial activity was observed. PLGA as positive control in short-term tests is not encouraged due to its limited biodegradability. Future work should consider longer durations, which may be necessary to provide sufficient biofilm colonization of rubber particles, and assess other (abiotic) degradation mechanisms.

**MO325 Evaluating sorption properties of tire materials using poly-parameter linear free-energy relationships (pplFER)**

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Tire materials are common representatives of microplastics in the environment. They are introduced on the one hand as tire wear, an abrasion product, which reaches the environment via road runoff. On the other hand recycled and shredded tire crumb rubber (TCR) is applied as filler material for example on turf fields. It was recently shown that tire materials are a substantial share (~ 66 %) on waste that is introduced into the environment as microplastic particles. Tires generally consist of a mixture of polymers (40-60 %), mostly styrene butadiene rubber (SBR). These elastomers are compounded with carbon black or silica as reinforcing agent (20-35 %), oils (15-20 %) as softeners and extenders as well as vulcanization chemicals (e.g. zinc oxide and sulphur (1-2 %)). Although tire materials are known to pose the risk of leaching toxic substances, they are one of the most popular construction materials and are widely distributed for example in the application of artificial wastewater treatment systems. Rubber may leach organic pollutants from water. The precise characterization of molecular interactions between tire materials and (organic) compounds is therefore important to evaluate and predict the behaviour of tire materials in aqueous systems. Poly-parameter linear free-energy relationships (pplFERs) provide the opportunity to describe the contributions of individual molecular interactions to overall sorption processes taking into account both the physico-chemical properties of the sorbate as well as the sorbent. They have been successfully used to describe and predict sorption of organic compounds to various sorbents. This work hence intends to investigate sorption properties of tire rubber crumb using poly-parameter linear-free energy relationships. [1] B. Liebmann, Mikroplastik in der Umwelt, 2015, [2] B. Bocca, G. Forte, F. Petrucci, S. Costantino, P. Izzo, Sci. Total Environ. 2009, 407, 2183; [3] C. Louthan, Microparticles: Occurrence, Effects and Sources of Releases, 2015, [4] Y. R. Lin and H. Teng, Microporous Mesoporous Mater. 2002, 54, 167. [5] R. B. Stone, L. C. Coston, D. E. Hoss, F. Cross, Mar. Fish. Rev. 1975, 37, 18.[6] L. Alamo-Noile, O. Perales-Perez, F. R. Roman, Desalin. Water Treat. 2012, 49, 296. [7] M. Abraham, A. Ibrahim, A. Zissimou, J Chromatogr A. 2004, 1037, 29. [8] S. Endo, P. Grathwohl, S. Haderlein, T. Schmidt, Environ Sci Technol. 2009, 43, 3094.  

**MO326 Particle toxicity in the daggerblade grass shrimp (Palaeomunida pugio): micronized tire wear particles and microplastics**

L. L. Halle, University of North Carolina at Greensboro / Biology; R. Leads, College of Charleston / Biology; S. Kell, College of Charleston / Graduate Program in Marine Biology; A.D. Gray, University of North Carolina at Greensboro / Biology

Recent surveys of Charleston Harbor, SC (USA) have demonstrated that >75% of total microplastics at some locations are tire wear particles (TWP). The aim of the present study was to investigate the toxicity of wet preserved TWP in adult grass shrimp (Palaemonetes pugio) and compare it to other microplastic particles. For our TWP assays, we conducted a 96-hour acute toxicity test and an immune challenge. Acute mortality was not observed at concentrations up to 100 g/L (1.9x10^17 particles/L). In our immune challenge, grass shrimp were exposed to TWP, polypropylene fragments, polyethylene spheres, polyester fibers, or sediment for 96-hours. Grass shrimp were then injected with either HEPEs-buffered saline or Vibrio comma bacteriophage. After 48 hours, no significant decrease in immune function was observed in exposed shrimp (p>0.8). We also conducted assays examining the size and shape dependent effects of microplastic particles (spheres, fibers and fragments), including TWP, on grass shrimp. Grass shrimp were initially exposed to various size fractions of plastic spheres (30, 35, 59, 75, 83, 116, and 165 µm), fragments (34 and 93 µm), fibers (34 and 93 µm), and TWP (50, 106, and 302 µm) at a concentration of 50,000 particles/L for three hours. Following exposure, grass shrimp were placed in particle-free water and monitored for survival, ingested and ventilated particles, and residence time in the gills and gut. Grass shrimp readily ingested and ventilated all tested particles. The time for mortality to be detected ranged from 18.7 to 24.2 hours up to 43±13.8 hours. Gut clearance for the TWP was 25.2±3.0 hours. Within the gill chamber the time for microplastics to be removed ranged from 27-45 hours with an average of 36.9±5.4 hours. Gill clearance for TWP was significantly longer at 51.2±21.2 hours. Mortality in these assays ranged from 0-55%, with microplastic spheres and fragments under 50 µm not acutely toxicity. All sizes of TWP were not acutely toxicity. Fibers were not toxic at both size fractions analyzed and increased porosity and roughness on rubber surfaces over the course of the experiments, seemingly indicating degradation via surface colonization. Overall, ready biodegradability tests proved suitable to obtain information on degradation of car tire rubber. This study provides first evidence of their degradability, especially for weathered rubber in the presence of a co-substrate, which should be considered for future detoxification assessment. Noteworthy, exposure of rubber to microbial activity was observed. PLGA as positive control in short-term tests is not encouraged due to its limited biodegradability. Future work should consider longer durations, which may be necessary to provide sufficient biofilm colonization of rubber particles, and assess other (abiotic) degradation mechanisms.

**MO327 Acute and chronic toxicity of micronized tire rubber to Hyalella azteca**

F. Khan, L.L. Halle, Roskilde University / Science and Environment; A. Palmqvist, Roskilde University / Department of Science and Environment

An average car tire lasts for 40000 km and during its life time 30% of the tire tread will be emitted into the aquatic environment. Inevitably it interacts with aquatic biota. In comparison to the wealth of research on the impacts of microplastics (MPs), there is little on micronized tire rubber (microrubber, MR). Recent reports suggested that tire rubber contributes a significant proportion of ocean’s plastic and when rubber is found in the environment it is often classed with MPs, but MR is fundamentally different from MPs in terms of structural and chemical properties, and perhaps should be considered as a distinct pollutant. MR contains a suite of toxic substances; trace metals (notably Zn, Cd), polycyclic aromatic hydrocarbons (PAHS), pyrene (a pyrene) and assorted volatile organics used in vulcanization and as antioxidants (e.g. aniline) which have been shown to leach into the aqueous environment. Currently, little is known about the ecotoxicological impacts of MR. The present study was to conceive to determine the toxicity of tire rubber particles to **Hyalella azteca**, an established freshwater model organism, over acute and chronic exposures, and to delineate particle effects from those of the leachate. The acute toxicity (48 h) of MR particles compared to the MR leachate show similar LC50s (3426±172 particles/mL for MR and 3628±672 particles/mL for leachate), but significant differences are found at LC10 and LC90, suggesting that at low particle concentration the leachate is more important, but at high concentrations the particle may act to deliver chemicals in vivo following ingestion. The results of the 21 day study showed that mortality, reproductive output (neonate production) and net growth were significantly impacted at the higher exposure concentrations of MR. MR is an emergent contaminant of concern that is similar but distinct to microplastics in many aspects. Very little is known about the toxicity of MR, but our results show that MR exposure has short-term and longer-term toxicity on a key freshwater species.

**MO328 Acute and chronic effects on Hyalella azteca and chemical analysis of rubber particles and leachate - comparison of pristine micronized car tire to previous data on worn car tire particles**

L.L. Halle, Roskilde University / Science and Environment; A. Palmqvist, Roskilde University / Department of Science and Environment; K. Kumpmann, A. Jensen, Danish Environmental Analysis; F. Khan, Roskilde University / Science and Environment

Microrubber (MR) from car tires constitutes a significant contribution to particulate microparticle debate. 30% of the tire tread is emitted into the aquatic environment during the lifetime of the tire. Mr rubber particles have been detected in the aquatic environment, the potential environmental impacts of this contamination are largely unknown. **Hyalella azteca** is an ecologically relevant freshwater amphipod that is also a well-established model organism in ecotoxicology. This study aims to investigate the acute and chronic effects of Microrubber (MR) on freshwater species. To determine particle effects and chemical characteristics of a pristine tire with previous data from a worn tire of same make and model. Effects are assessed as changes in survival, growth and reproduction and both the effects of rubber particles and rubber leachate is investigated with the aim of determining whether there are particle effects and/or if the mode up uptake of chemicals leached from tire influences effects observed in unicellular. The chemical characteristics of both pristine and worn tire particles are quantified by GCMS. The preliminary results indicate that, surprisingly, pristine tire both as particles and leachate is much more toxic than worn tire in acute tests. Although the main source of MR undoubtedly is worn tire, these results points toward further ecotoxicological testing of tire coatings used during manufacturing.

**MO329 Applying nuclear techniques to study the biokinetics and toxicodynamics of**

R. Lin and H. Teng, University of Vienna / Environmental Geosciences; Nole, O. Perales Charleston / Biology; R. Leads, College of Charleston / Biology; A.D. Gray, University of North Carolina at Greensboro / Biology

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microplastics and co-contaminants in marine biota
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Despite recent efforts in understanding the risks associated with marine plastic pollution, there remains a great deal of uncertainty regarding the potential impacts of microplastics and nano-particles on wildlife and humans. This largely relates to the methodological and analytical limitations associated with studying relatively low and environmental concentrations of these plastics. The IAEA Radiocology Laboratory, in collaboration with a team of external experts, is tackling these challenges by applying nuclear and isotopic techniques to address important outstanding questions on the risks of microplastics to marine organisms. Novel approaches using radiolabeled plastic particles and associated organic and inorganic contaminants are being developed to very precisely quantify their movement, fate and impacts on a range of aquatic biota, under controlled laboratory conditions. Nuclear techniques are uniquely suited for this research given their sensitivity and capacity to measure biokinetic and toxicodynamic parameters over time. As such, these tools will allow us to address important knowledge gaps, including: (1) the biokinetics, bioaccumulation and incorporation of their biological impacts of realistic concentrations of small plastic particles (< 100 μm) in marine biota; (2) the sorption kinetics of trace pollutants to microplastics; and (3) the influence of microplastics on the bioaccumulation of co-contaminants. Importantly, this research will allow us to test if microplastics can truly be bioaccumulated (i.e., cross epithelial membranes/tissues), and if they can act as a vector for contaminants in the marine environment under low exposure conditions. This poster will provide an overview of the techniques used to address these questions, as well as preliminary outcomes and future directions.

M3030 Aggregation kinetics of plastic nanoparticles in fresh and marine phytoplankton culture media
H. van Epen, EPIC, University of Bordeaux / UMR EPOC 5805; J. Gignault, University of Rennes 1 / Laboratoire de Geosciences Rennes; M. Baudrion, Université Bordeaux 1 / UMR EPOC 5805

Release of plastics debris in the environment has been catching more and more concern in recent years, especially in aquatic environment. It has been observed recently, that plastics break down to produce nanoparticles by photochemical degradation in marine waters. However, there is a lack of suitable analytical tools, and the environmental fate and transport mechanisms of nanoparticles have not yet been investigated. Indeed, several ecotoxicology studies investigate the impact of nanoparticles on aquatic organisms without addressing their aggregation state in aqueous media, whereas the (1) the biokinetics, bioaccumulation and incorporation of their biological impacts of realistic concentrations of small plastic particles (< 100 μm) in marine biota; (2) the sorption kinetics of trace pollutants to microplastics; and (3) the influence of microplastics on the bioaccumulation of co-contaminants. Importantly, this research will allow us to test if microplastics can truly be bioaccumulated (i.e., cross epithelial membranes/tissues), and if they can act as a vector for contaminants in the marine environment under low exposure conditions. This poster will provide an overview of the techniques used to address these questions, as well as preliminary outcomes and future directions.

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M3031 Mytilus spp. as sentinel species for water borne microplastic ingestion; a case study from the Norwegian coast

Mytilus spp. is widely used as a sentinel species for coastal pollution monitoring, and ICES have suggested M. edulis and M. galloprovincialis as suitable to also monitor waterborne microplastic pollution in the marine environment. Due to several methodological differences between studies (e.g. sample collection, samples processing identification of microplastics such as visual ID, detection limit and chemical verification of polymers) comparability can be challenging. When looking at spatial trends it is necessary to use comparable methods. Several studies have found microplastics in Mytilus spp. and a recent study found accumulating levels of microplastics in mussels compared to the surrounding waters. In this study, a total of 252 mussels were investigated from 13 different sites along the Norwegian coast by using KOH digestion followed by visual ID and µFTIR. Occurrence of plastics were found in a total of 76.6% of the analysed mussels from the Norwegian environment, with the overall average plastic load being 1.85 particles gram⁻¹ w.w (ranging from 0 – 24.45) and with the highest values found in mussels from the Barents Sea. Microplastics consisted of fibers (85 %), fragments (10 %) and foams (5 %) and most particles were 1 mm, with the most dominant polymer group being semi-synthetic materials (rayon/viscose). Based on literature and this current study, Mytilus spp. seems to be a promising sentinel species to monitor pollution of the smallest waterborne microplastics in the marine environment. However, for the method to be fully quantitative, improvements/further investigations are needed e.g. same sized mussels from each site using a standardisation in the sampling and processing methodology within populations. Furthermore, the presence of semi-synthetic materials in the marine environment needs to be further investigated as well as their potential effects on biota.

Mercury Biogeoosciences - Fate, Effects and Policy (P)

MO333 Influence of biofilm composition on mercury bioaccumulation

In aquatic systems, the dominant lifestyle of microorganisms (bacteria and microalgae) is to live together interlocked in exopolymeric substances (EPS), rather than as single cells. Besides their involvement in several biogeochemical processes, biofilms are sites of accumulation and transformations of mercury (Hg). Their natural assemblage of heterotroph and autotroph microorganisms makes them an important entry of Hg into aquatic food webs. The objective of the present study is thus to better understand the mechanistic processes that control Hg accumulation in biofilms and we focused on the elucidation of the role of biofilm composition on the kinetics of Hg uptake. For that end, two couples of biofilms were exposed to HgI₂ (~100 pmL, precisely measured) using microcosms. The first couple of biofilms was obtained using different times of colonisation e.g. 85 and 127 days whereas the second couple (bottom/surface biofilms) was obtained owed to different colonisation depth in the Versoix River (CH). Prior Hg exposure, biofilm biomass and microbial composition (chlorophyll content and transcriptional abundance of 16S rDNA gene) was determined as well as EPS and mineral EPS thiol content. The main water quality parameters (pH, concentrations of dissolved organic carbon, Hg, anion and cation) of the exposure media were also analysed. Accumulation of total Hg and non-extractable Hg (determined after a cysteine washing step) in biofilms were measured at different step times (t < 24 h) to model non-extractable Hg uptake kinetics using a non-linear pseudo first order one-compartment model. In each biofilm, non-extractable Hg accumulation was very rapid, within minutes of exposure. The uptake rate constant of the younger biofilm was measured to be 10 times higher than that of the older biofilm. That same ratio was also obtained between the bottom and the surface biofilms. Except for the older biofilm, Hg accumulation reached a plateau at ~6 Hg exposure. A decrease in the EPS thiol concentration was observed in the bottom biofilm upon Hg exposure, suggesting a change in Hg bioavailability in the microorganism environment living in that biofilm. Our study demonstrated that biofilm microbial and EPS composition as well as thickness influence Hg uptake by microorganisms living in biofilms.

MO334 Gaseous elemental mercury concentration and diurnal evasional fluxes from the water-air interface in coastal environments of the northern Adriatic Sea
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Among pollutants widespread in the environment, mercury (Hg) has been recognised for its toxicity, mobility and bioaccumulation potential. In coastal areas these presence of this element generates conflicts with important resources of profitable value such as fisheries and aquaculture. The Marano and Grado Lagoon (Adriatic Sea) experienced a double Hg impact. The first is due to the mining activity second is the result of discharge of a chloralkali plant effluents. Fish farming is a METIAN, University of Bordeaux / UMR EPOC 5805; J. Gigault, University of Geneva / Département F. A. Forel des sciences de l’environnement et de la santé; S. Le Faucheur, Institute F.-A. Forel, University of Geneva / Département F.-A. Forel des sciences de l’environnement et de la santé
historical activity covering 14% of the total lagoon area. Recently, one fish farm was long-term monitored in order to understand the role of the sediment-water interface in recycling Hg and to estimate benthic fluxes and Hg mobility in the water column. An important further step toward a better comprehension of the Hg biogeochemical cycling in the lagoon environment, is represented by the estimate of its evasion fluxes, as gaseous elemental mercury (GEM), at the water-air interface. A combination of sediment pore water mercury concentrations from a high-mercury area (Lumnes-RA 915+) has been used to measure GEM and to estimate the diurnal evasion flux at the water-air interface during three seasonal campaigns in four selected sites: two in a lagoon fish farm, one in the open lagoon environment highly impacted by Hg mining activities and the last one in an uncontaminated area of the Gulf of Trieste, the Bay of Piran (Slovenia). Accomplished to these missions is the role of a background level of gaseous elemental mercury observed in biotic media and vulnerable populations on the basis of available scientific, environmental, technical, financial and economic information. UN Environment in close collaboration with Italian National Research Council - Institute of Atmospheric Pollution Research (CNR-IAR) and WHO implemented a UN Environment Global Environmental Facility (GEP) project entitled “Develop a plan for global monitoring of Human exposure to and environmental concentration of Mercury”, from late 2014. The project contributes to scientific knowledge for development of effectiveness evaluation mechanism. The main aim of the project was to harmonize approaches for mercury monitoring and to strengthen the capacity for mercury analysis in humans and in the environment. The experience made within the project suggests that there is a urgent need to coordinate the global efforts in atmospheric mercury monitoring by integrating existing monitoring programs worldwide in cooperation with other on-going programs such as GEO (Group on Earth Observation, www.earthobservations.org) and specifically with the GEO Flagship “Global Observation System for Mercury (GOS-Mercury)”. That observation is especially true when contaminants are metals, notably Hg, which bioaccumulates in fish (in gills, muscle, gonads and liver). Results coming from the pilot survey campaigns carried out with mercury novel biovector for metals, which biomagnifies in food webs (Gochfeld 2003). Mercury concentrations in sediment and bryophytes are low, with no observable differences between up- and downstream sites. However, fish gills, liver, and muscle demonstrated elevated Hg concentrations downstream of sites compared to upstream ones. In fall as well as in early-spring, when the flow decreased by more than half as compared to that of summer, Hg concentration in the water column is lower but concentration in fish and bryophytes, as well as Hg concentrations in are in fish and algae, significantly increased the Hg increase downstream the landfills. However, no oxidative stress and impairments are observed in fish. The present study confirms the need to address all compartments to properly assess the water quality of an aquatic system and therefore to understand potential impact of landfills and industrial sites on freshwater ecosystems.

M0337 Mercury Photo-reduction and Total Photoreducible Mercury Dynamics in the Lakes of Kejimkujik National Park, Nova Scotia N.J. OD’Riscoll, Acadia University / Department of Earth and Environmental Science; J. Kickbush, Acadia University / Biology; N.J. O’Driscoll, Acadia University / Engineering; S. Klapstein, Acadia University / Earth & Environmental Science; A. Loder, Acadia University / Natural Resources; S. Klapstein, Acadia University / Biology; M.L. Mallory, Fern Hill; N.J. OD’Riscoll, Acadia University / Department of Earth and Environmental Science

Mercury is a persistent and bioaccumulative chemical that is present in many remote environments due to its ability to be transported long distances in the atmosphere, and to be deposited far from the original source (Sunderland and Chnura 2000). Wetland ecosystems are important “hot spots” for mercury in eastern Canada, providing anoxic environmental conditions that promote the bacterial methylation of mercury. Methyl mercury is the most biologically available form of mercury and the form which biomagnifies in food webs (Gochfeld 2003). A mercury biovector for methyl mercury – benthic invertebrates – including mercury-bound nutrients, which may indirectly affect metal speciation (Choy et al. 2010). The site for this study, Big Meadow Bog (Brier Island, Nova Scotia, Canada) has a history of ditching in the 1950s, which changed hydrology significantly, resulting in colonization by 3000 pairs of herring gulls (Larus argentatus) in the 1980s. To quantify changes in mercury mobilization and speciation in response to this disturbance, groundwater samples from a reference bog with similar geological and hydrological characteristics. The filtered samples were analyzed for total mercury, methyl mercury, and water chemistry (pH, conductivity, anions, cations, and dissolved organic and inorganic carbon). Results show significantly higher nutrients (nitrate, phosphate, and sulfate), total mercury, and methyl mercury concentration when compared to the reference bog that is minimally impacted by avian biovectors. This elevated availability of methyl mercury could potentially pose a threat to the local ecosystem and wildlife population due to mercury’s toxicity to living organisms (Akearok et al. 2010, Singh et al. 2011). Citations: Akearok J et al. 2010. Science of the Total SETAC Europe 28th Annual Meeting Abstract Book
Organohalogen and mercury residues in fish from the Western Mediterranean Sea: concentrations, bioaccumulation and dietary exposure
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Organochlorine compounds (OCs) and mercury (Hg) have diverse deleterious health effects and are persistent in the environment. They tend to bioaccumulate and biomagnify along the food chain. Diet is the major source for the incorporation of these pollutants into humans, especially through consumption of fish. This research focuses on the presence of OCs, total mercury (THg) and methylmercury (MeHg) in lean fish from the Western Mediterranean Sea. Determinant factors of these concentrations such as trophic level and weight of fish have been studied. The estimated weekly intake (EWI) from the Spanish population of these pollutants has been assessed. Samples were collected between March 2015 and August 2016. Most of them were from Balearic Islands (Majoreca n=67, Menorca n=17, and Ibiza n=18) and the rest of the samples were from Tunisia (n=2) and Egypt (n=1). Additional fish samples were collected from the Atlantic Ocean, in front of Senegal (n=4) and Mauritania (n=10) coasts for comparison. The OCs levels found in fish were similar or lower than in other previous studies. In contrast, 15% of the most frequently fish species consumed by the Spanish population had Hg concentrations above the maximum level set forth by the European Union MRL for human consumption. The concentrations of OCs and Hg between trophic levels did not differ significantly. Except for HeCB, the values were higher in the upper trophic level with statistically significant differences for ΣDDTs and ΣPCBs (p < 0.05). The relationship between fish weight and pollutant concentrations were also studied. A positive relation between Hg concentrations and weight was found (R2=0.58; p-value < 0.001). This trend was not observed for any other OC. The concentrations found in dusky grouper from the Mediterranean Sea and Atlantic Ocean were also compared. The former group presented higher levels for ΣDDTs, ΣPCBs and Hg (p < 0.05). The estimated weekly intake of OCs were well below the reported Tolerable Intakes. However, for Spanish population that only consume Mediterranean fish, the estimated weekly intake for Hg (4.42 µg/kg bw) exceeded the threshold of 3 µg/kg bw established by EFSA in 2012. 4 µg/kg bw being considered as a safe level for MeHg, involving provisional weekly tolerable intakes of 1.3 µg/kg bw were six and three times higher than these provisional tolerable weekly intakes in adults and children (7-12 years of age), respectively.

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Fish consumption is linked to the prevention of some human diseases, especially reduced risk of cardiovascular and related neurological disorders, due to the content of high-quality protein, vitamins and n-3 fatty acids. At the same time, fish consumption is considered a major pathway of mercury (Hg) exposure in human. More than 90% of this Hg present in fish tissue is found essentially in its organic form (methylmercury (MeHg)), which is the most toxic form of Hg. Due to the potential adverse human health effects, international agencies have established Reference doses (RfD) as recommendations to Hg intake. Some studies have been associating the fish consumption habits, portion sizes and other characteristics of the population of the Metropolitan area. Shark mercury content was taken from the previous studies, rather than a result of increasing availability of mercury to fish. A previous three years study of mercury content in a variety of edible marine fish from sea bass, catfish, tilapia, red snapper and other populations. Compilations by others between the first two studies (1980 and 1998) had suggested an increasing trend of mercury in fish. Assessment of the mercury concentrations in fish caught in 2015 against those reported in the previous studies found that the concentrations have remained relatively stable from 1980 to 2015, regardless of the location from which fish were collected. There was no indication of increasing concentrations of mercury over time with the exception of the red snapper. While some variation was observed in the levels of mercury in fish between studies, this variation appeared to be due to differences in the size and age of fish between studies, rather than a result of increasing availability of mercury to fish.

Mercury health risks due to the substitution of fish meat with shark meat.
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A previous three years study of mercury content in a variety of edible marine fish from sea bass, sea bream, catfish, tilapia, red snapper and other populations. Chondrichthyes universal oligouconicoleotides in PCR were used to analyze the samples. 777 surveys were applied to obtain information regarding fish consumption habits, portion sizes and other characteristics of the population of the Mexico City metropolitan area. Shark mercury content was taken from the previous study. Hg average daily dose, lifetime average daily dose and total dose were calculated for two concentrations: 0.13 mg/kg and 0.13 mg/kg (mg Hg/Kg). Health risk was calculated using USEPA equations. Of the 52 “fish samples” analyzed 61.53% were identified as sharks of the following species: Leopard (Galeocerdo cuvier), Common sawshark (Pristiophorus cirratus), Goblin (Mitsukurina owstoni), nurse (Ginglymostoma cirratum), whale shark (Rhincodon typus), scalloped hammerhead (Sphyraena lewini), dagger-nose (Isogomphodon oxyrhynchus), silky (Carcharhinus falciformis). With regards to the health risk, when considering the lowest Hg concentration, children may eat only one 188 g portion/month, while fertile women and older people, two 190 g portions/month and men up to five 260 g portions/month. When considering the average Hg concentration, the number of portions/month is drastically reduced to less than one portion/month. If the amount of portions described before are respected, the calculated risk for babies was 0.83 but significantly reduced later in life if the lowest Hg concentration is considered. However, when calculating the risk with the average Hg concentration the risk quotient was always above 1 and up to 7.2 for babies. In conclusion there is a health risk when eating fish that cannot be identified, so people must buy whole fish to secure their identity and authorities should implement a monitoring program to penalize the sale of shark meat as fish meat.

Mercury in trophic webs of estuaries in South-Eastern Brazil.
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The estuarine regions of Brazil are susceptible to anthropic pressures due to urban, industrial and agricultural activities. Despite low Hg levels in the region, there exist knowledge gaps related to Hg bioaccumulation in marine food chains. In this paper, the authors present a synthesis of data on Hg concentrations in estuarine fish from the Southern Brazil. This data was obtained from 2005 to 2018, with a focus on fish species from the states of São Paulo, Paraná, and Rio de Janeiro. The authors found that Hg concentrations were higher in fish species from the Atlantic coast compared to those from the Pacific coast. They also observed a positive correlation between Hg concentrations and weight of fish, with a coefficient of determination (R2) of 0.58. In addition, they reported that Hg concentrations were higher in the upper trophic level compared to the lower trophic level. The authors concluded that further research is needed to better understand the factors influencing Hg bioaccumulation in marine food chains and to develop effective strategies to mitigate Hg pollution in estuarine regions of Brazil.
industrial, agricultural and harbor activities, which impact the environment through the release of contaminants such as metals. Among these metals, mercury (Hg) is highlighted due to its toxicity and capacity of biomagnification. In the Paranaguá estuary, in the state of Paraná, and in the Cananéia estuary, state of São Paulo, these potentially polluting activities are present in different levels. However, these regions comprise the largest remnants of the Atlantic Rainforest ecosystem, fact that gives them the status of World Heritage site and biodiversity hot spot (UNESCO), therefore a environmental monitoring and conservation acts of the areas are necessary. Thus, this study investigated the concentrations of Hg and nitrogen isotope ratio ($\delta^{15}$N) in the trophic web composed by benthic invertebrates, benthivores fish (Stellifer rastrifer, Paralabrus brasiliensis and Isopisthus parvipinnis) and marine mammals (Sotalia guianensis and Pontoporia blainvillii), to test and the trophic dynamics of Hg and compare these estuaries. Samples were taken in the summer of 2015; analyses of $\delta^{15}$N were carried out by Elemental Analysis - Isotope Ratio Mass Spectrometry (EA-IRMS), and mercury analyses were carried out by Optical Emission Spectrometry, Inductively Coupled Plasma with Vapor Generator Accessory (OES-ICP-IVA), in the muscular tissue of the organisms. The results of $\delta^{15}$N varied from 6.4 to 13.8 % in Paranaguá and from 7.1 to 14.3 % in Cananéia, with continuous enrichment among the trophic levels. Concentrations of Hg were significantly higher in Paranaguá (0.02 to 5.8 mg kg$^{-1}$) than in Cananéa (0.02 to 0.9 mg kg$^{-1}$), with maximum values in marine mammals, followed by invertebrates benthic and fish. Through linear regressions between Hg and $\delta^{15}$N, positive correlations were observed only in Paranaguá, but they were not significant, which indicates a trend of biomagnification of this element. Such a behavior is expected since the input sequences of Hg can accumulate from anthropic activities than Cananéa, and presented similar values to highly degraded Brazilian estuaries, such as Santos Bay (SP) and Guanabara Bay (RJ). The results showed a current panorama of the trophic distribution of Hg in these estuaries being a useful tool in environmental monitoring and coastal management in regions of great ecological importance.

MO344 Biological and Geochemical Drivers of Mercury Toxicity in Yellowknife, NWT, Canada
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MO345 Use of green tea to reduce mercury and methylmercury bioaccessibility in raw and cooked fish
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MO346 Importance of a tidal flat-saltmarsh system as a source-sink of mercury in a contaminated coastal lagoon environment (northern Adriatic Sea)
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Saltmarshes are important constituents of marine-coastal transitional environments that provide several services to the ecosystem. Due to anthropogenic activities, several contaminants are increasingly introduced into these environments where are trapped by saltmarshes vegetation. Bioaccessibility of these contaminants and, consequently, their availability can be influenced by several factors such as, for example, saltmarshes vegetation and its interaction with water. The use of saltmarshes as tools in environmental monitoring and coastal management is a well-recognized approach. This work aims at determining the potential role of the tidal flat-saltmarsh (TF-S) system as a sink or secondary source of Hg in this coastal lagoon in relation to the chemical-physical processes leading their remobilization. The main objective was to understand the role played by periodic flow of tide in a TF-S pilot site in terms of mercury bioavailability and accumulation and release of Hg. Tidal flows and water chemistry were measured at the mouth of a principal tidal creek which collect the waters of a dense channel network draining a 5.5-ha tidal flat-saltmarsh system. Tidal fluxes were estimated by combining discrete hourly tidal flow measurements with weighted measurements of particulate (PHg) and dissolved (DHg) mercury obtained by water samples. The highest values of DHg and PHg were recorded during ebb tide and the lowest values of fluxes estimated in flood tides. These results, from the preliminary period of sampling, confirmed other evidence of serious morphological deterioration of this coastal environmental. Keywords: tidal flat-saltmarsh system, mercury, tidal fluxes, sedimentary budget.
The net production of MeHg is controlled by both mercury methylation and the product of illegal mining. Using a direct quantification method (EPA 7473), it was studied the variation of total mercury (HgT) in samples of the riparian fern Thelypteris hispidula, sediments and water in three streams: El Socorro, El Rato, and El Pato, during the dry season, dry-rain transition and rainy season. Using non-parametric statistics (Kruskal Wallis), significant differences were found in the HgT concentrations in the samples. HgT was on a spatial level in each studied matrix (p < 0.05), accumulating mainly in El Socorro. The HgT in the root of the plants presented differences in distinct sampling times (p = 0.005), increasing in the rainy season. The Spearman’s bivariate correlations showed that the dynamics of HgT accumulation in the root, is directly related to the concentration of HgT in the stem (rho = 0.918, p = 0.000) and leaves (rho = 0.900, p = 0.000). It was also evidenced that the accumulation of HgT in the root, is influenced by the concentration of HgT in the sediments (rho = 0.764, p = 0.001). These results demonstrate the environmental effects caused by mining activities in protected areas in Colombia.

M0349 Temporal integration of diurnal variations of metals and mercury concentrations by passive sampling method in a highly polluted site on the Deûle River, northern France

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Due to several metallographic plants along the river, the Deûle River is a highly polluted site in the northern France. Previous studies showed that sediments are polluted with Cd, Pb, Zn and Hg (Vdovic et al., 2006). Moreover, intense fluvial traffic generates regular resuspension of sediments leading to diurnal variations of metals concentrations in the dissolved and particulate phases. This was previously highlighted by in situ voltmeter approaches allowing to measure Pb, Zn and Cd with a high frequency data acquisition (Superville et al., 2014). However, these measurements could not be performed for Hg with such probes, while previous field deployment of DGT (Diffusive gradient in Thin films) passive sampler suggested such variations. Indeed, time weighed average concentrations were 20 times higher (20 ng/L) than those measured in grab samples (≈ 1 ng/L). Thus, a field campaign was conducted during 15 days to assess diurnal variations of metals and Hg concentrations (dissolved, particulate and labile phases) related to fluvial traffic in the Deûle River, in the city of Aubry, downstream a metallurgical plant (Umicore). During the first week, DGT were exposed and grab samples were collected with a high sampling frequency (night and day). Then, a second set of DGT were exposed during the second week. The resuspension of particles and anoxic sediment caused by fluvial traffic was highlighted by the simultaneous increase of metal and Hg concentrations in DGT, to 0.70 µg/L (3 to 34 mg/L). The analysis of metals and Hg in SPM showed increases of Pb, Zn, Hg and Cd concentrations in the particulate phase. The analysis of particulate Hg after two different filtrations at two cut-off points (0.45 and 0.70 µm) showed that particulate Hg re-suspended by fluvial traffic was mostly in the coarse fraction of SPM.

Furthermore, the increase of SPM concentrations was related with an increase in dissolved Hg concentrations. These results suggest that when the anoxic sediment is remobilized by barge traffic, particles could be oxidized inducing a release of Hg. Moreover, since changes in redox conditions could also induce modifications in Hg speciation, further analysis will be carried out to measure Hg^0 and CH3Hg^+.

Finally, the interpretation of DGT measurements will show how well DGT integrate variations of inorganic contaminants concentrations during the exposure period.

M0350 The effect of activated carbon amendment on mercury methylation in contaminated sediment

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The conversion of elemental and inorganic mercury (Hg) to monomethyl mercury (MeHg) has been widely studied due to MeHg being more bioavailable, bioaccumulative and toxic to humans than the inorganic and elemental species. The net production of MeHg is controlled by both mercury methylation and demethylation, and a range of factors affects both processes. Sediments are known sites for MeHg production, as they are sinks for Hg, they have suitable red-ox conditions, a presence of methylaing bacteria and more. Activated carbon has been much explored as a remediation tool for contaminated sediments. The carbon can immobilize contaminants, inhibiting the release to the water column and uptake in biota. Studies have shown that activated carbon also adsorb Hg-species, but the mechanisms of how a carbon amendment affects the processes of Hg-methylation in contaminated sediments is not well understood. A lab trial was set up with sediment from two sites in Norway: The Gunnekleiv fjord (GF) and Bergen Harbour (BH) – two heavily contaminated locations. The sediment was sieved using DGTs with an aragonite diffusion gel and a spheroid-thion resin gel. Pore water data show a net production of MeHg in the GF control, from an initial 8.7 to 393 ng/L within the first month, but it then drops off to 147 and 18.4 ng/L after 3 and 6 months respectively. Compared to the control, an initial reduction of 86% MeHg in pore water is seen for the AC treatment, that increases to >95% for the 1, 3 and 6 month time series. The BC treatment caused an initial 55% reduction of MeHg but after 1, 3 and 6 months the reduction is >99% compared to the control. In the BH sample, there was no increase of the initial 2.1 ng/L MeHg in the pore water of the control during the 6 months of the experiment. Both AC and BC treatments however, reduced MeHg in the pore water by >50%. Pore water MeHg-concentrations measured by DGT were similar to concentrations in extracted pore water, indicating that pore water MeHg is available for uptake.

M0351 Bayesian Human Health Risk Assessment of Almaden Mining Area

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Almaden, with the largest and richest known mercury deposits is located in the southeast of Ciudad Real (Spain) with a population of 5,657 inhabitants (2016). This area can be considered one of the most affected by mercury in the world, both by its natural origin and by anthropogenic pollution since there are indications that the cinnabar mines of this region have been mined without interruption since before the fourth century BC until 2002. A probabilistic human health risk assessment has been carried out in order to establish whether mercury contamination of Almaden environment endangers human health or not. A Probabilistic health risk assessment was performed to evaluate the exposure to mercury in a priori determined distributions that allow better estimation of the risk. The results show that the human health risks obtained by ingestion of fish bought in local markets and vegetables grown in the area (lettuce, beans, tomato, onion, pepper, potato, cucumber and zucchini were analyzed) are not acceptable. The probability analysis is that it is not always easy to obtain distribution functions for the different parameters of a given population, being frequent to use the literature to be able to complete the necessary information. To address this problem, Bayesian statistics have been used. Thanks to that, a combination between stablished density functions (priori distributions) and data collected at the study site can be carried out. In this way, the exposure variables are better defined by a posteriori-determined distributions that allow a better estimation of the risk.

In this way, the exposure variables are better defined by a posteriori-determined distributions that allow a better estimation of the risk. The results show that the human health risks obtained by ingestion of fish bought in local markets and vegetables grown in the area (lettuce, beans, tomato, onion, pepper, potato, cucumber and zucchini were analyzed) are not acceptable. The difficulty of the probabilistic analysis is that it is not always easy to obtain distribution functions for the different parameters of a given population, being frequent to use the literature to be able to complete the necessary information. To address this problem, Bayesian statistics have been used. Thanks to that, a combination between stablished density functions (priori distributions) and data collected at the study site can be carried out. In this way, the exposure variables are better defined by a posteriori-determined distributions that allow a better estimation of the risk.
also exists in oxidised forms (i.e. Hg(II)) that are reactive and can be transformed into organic Hg species such as methylmercury (MeHg), the most toxic Hg species and the one most prone to bioaccumulation in aquatic systems. Half of atmospheric Hg emissions are of natural origin whilst the rest are of anthropogenic sources, primarily from fossil fuel burning and other high temperature industrial processes (cement clinker production, waste incineration, ore roasting, steel production). Knowledge of Hg speciation both in air and in stack gas emissions is critical when validating models for predicting Hg emissions, transport, deposition and fate at the European level as well as on a global scale. Therefore, atmospheric Hg isotopic signatures that can be used to trace the origin and fate of atmospheric Hg also need metrological support and development. The overall goal of the EMPIR – MercOx project (Oct 17 – Sept 20) is to develop SI traceable measurements, for monitoring and control of mercury and its different species in gas emission sources and in the atmosphere. The project will achieve significant improvements in the measurement comparability and uncertainty of Hg measurement results. Currently, traceable calibration methods only exist for elemental mercury, but such measurements are also needed for oxidised Hg species in order to meet the requirements of EU regulation and the implementation of the Minamata Convention. The development of reliable and direct Hg(II) measurement techniques and reliable and traceable Hg(II) standards is needed to solve the traceability problem that currently exists in the measurement of total mercury (Hg(0)) and oxidised Hg concentrations originating from different Hg sources. Furthermore, methods for measuring oxidised Hg and for accurately comparing the Hg(II) concentration in generated elemental and oxidised Hg reference gas standards are required, as well as issues/organ-specific burden of Hg(II) and mortality. A four-parameter Hill model was used for the on-line measurement of Hg under field conditions and a comparison of Hg species inter-conversion.

MO354

PBTK/TD assessment of mercury (Hg(II)) accumulation in freshwater tilapia species

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Waterborne and dietborne exposures of freshwater fish to inorganic mercury (Hg(0)) affect their growth and reproduction. However, a mechanistic model to predict the impact of Hg(II) on freshwater fish is lacking. The purpose of this study is to develop a physiologically-based toxicokinetic/toxidynamic (PBTK/TD) model to assess bioaccumulation of Hg(II) in freshwater tilapia. A PBTK model consisted of six interested compartments can be constructed including blood, gill, liver, kidney, intestine and whole body. The elemental and oxidised mercury and physicochemical parameters can be estimated from published tilapia-related studies. The partition coefficients were estimated for each tissue or organ based on the experimental data by dividing Hg burden in tissues of that in blood at specific days after Hg(II) exposure. A series of experimental data were analyzed to reconstruct the dose-response profiles describing the relationships between tissue organ burden and Hg(II) and mortality. A four-parameter Hill model was used to describe the dose-response relationships. Here we showed that tissue/organ burdens would reach equilibrium before 180 days of exposure in all six rivers. Among all exposed tissues/organs, kidney had the highest internal exposure doses of Hg(II) ranging from 0.0208 – 0.1348 µg g⁻¹ ww. In contrast, muscle had the lowest internal exposure doses of 0.0001 – 0.0003 µg g⁻¹ ww(Hg(II)), indicating that Hg(II) was in muscle may be well below that required at risk for human consumption based on regulation from Taiwan FDA. The highest accumulative internal dose of Hg(II) was in gill of 0.0115% (CF: 0.0007 – 0.1907). The effective Hg(II) burden in tissue/organ at 50% mortality for liver, gill, and muscle were 10.410 ± 1.047, 6.307 ± 0.756, and 2.839 ± 0.575 µg g⁻¹ ww, respectively. A fair quantitative agreement between model predictions and experimental data was also reached. Sensitivity analysis indicated that the amount of Hg accumulated in tilapia whole body was most influenced by sediment uptake rate, indicating that sedimentborne Hg exposure was the most influential factor on accumulation of tilapia that is bottom-feeding fish. We suggest that more dose-response data of sublethal and chronic effects are required to improve future risk assessment in a mechanistic and practical way. In a broader way, our model can be applied to predict continuously chronic Hg accumulation in fish that are deemed safe for human consumption.

MO355

Mercury in fish, fish intake and fish consumption recommendation

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Fish consumption is recognized as an important component in the human diet, due to the high-quality protein content and n-3 fatty acids, however, it is also considered the main route of mercury (Hg) exposure from ingestion of contaminated seafood. Hg is released into the environment by natural and anthropogenic sources and is recognized as a pollutant of high importance, due its high degree of toxicity, persistence and bioaccumulative properties. Since exposure to mercury poses human health at risk, the Food and Agriculture Organization (FAO)/World Health Organization (WHO), Joint Expert Committee on Food Additives (JECFA) and also by the United States Environmental Protection Agency (USEPA) have been established reference doses (RfD) or “Provisional Tolerable Weekly Intake” (PTWI), in order to minimize that risks. JECFA established a PTWI for MeHg of 1.6 µg kg⁻¹ bw⁻¹ week⁻¹, whereas USEPA pointed a lower value of MeHg intake, setting the RfD at 0.14 µg kg⁻¹ bw⁻¹ day⁻¹ (equivalent to 0.7µg kg⁻¹ bw⁻¹ week⁻¹). Recently (2012), PTWI suggested by JECFA for MeHg was revised by the Environmental Food Safety Authority (EFSA) to 1.3 µg MeHg kg⁻¹ bw⁻¹ week⁻¹. This study, presents a brief review of the Hg concentration present in the muscle of commercially valuable fish species caught near the mid-Atlantic ridge (Azores Archipelago) where fish consumption e relatively high (……) and compares these Hg concentrations with the maximum levels of Hg for certain contaminants in foodstuffs established by the European community, evaluates the human exposure to Hg, using the Hg concentration quantified in scalp hair and related with fish consumption using a food frequency questionnaire and establishes isocurves pointing the maximum number of fishmeal per week without exceeding the MeHg RfD (USEPA RfD), by combining number of meals (per week), amount of fish ingested (by meal) and levels of MeHg in fish. The Hg concentration found in the hair indicates that individual with higher fish consumption per week generally have higher concentrations of Hg and in order to meet the USA dietary guidelines, which recommend a consumption of 227g; only fish with MeHg concentrations below 0.34 µg g⁻¹ could be selected to be consumed so that the PTWI established by JECFA would not be exceeded, despite the concentration of 0.5 µg g⁻¹ (for most of the fish species) or the concentration of 1.0 µg g⁻¹ (“exception list”) is allowed for fish consumption.

Mechanistic effect modelling for risk assessment: applications, use in a regulatory context and future directions (P)

MO356

Ring-test of different implementations of the General Unified Threshold Model of Survival (GUTS)

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The additional information and insight gained through the application of toxicokinetic-toxicodynamic modelling can strengthen the environmental risk assessment of chemicals in consumer products or plant protection products. For the endpoint survival the most suitable and powerful tool is currently the General Unified Threshold model of Survival (GUTS), which consists of two complimentary models: GUTS-SD (stochastic death) and GUTS-IT (individual tolerance). In order to ease the use of GUTS and increase trust and acceptability we recently carried out a ring-test of eleven different implementations of GUTS. The frequency of erroneous results due to programming errors and less appropriate settings for numerical solvers or parameter search and convergence algorithms indicated that user training and experience is key. However after correction of user errors all software implementations resulted in comparable and similar results. Estimated parameter values generally agreed well and the implementations returned similar results across scenario and threshold mixtures. The results suggest that new users and new GUTS implementations should be trained using this ring-test and refer to these results as benchmark. Any new user should run the ring test exercises and improve their modelling techniques until they achieve comparable results. Standardisation of typical use cases could also help to reduce sources of error as well as corresponding, user-friendly, robust GUTS software. This software could reduce sources of error by restricting user-choice to only those options suitable and relevant for the regulatory risk assessment under consideration.

MO357

Feeding impairment in fish explained by a TK-TD model

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Ecotoxicology In current environmental risk assessment (ERA), plant protection products (PPPs) are tested on a diversity of standard test species for harmful effects. Recent developments in mechanistic effect modelling provide the possibility to extrapolate results from standard studies to unknown species or unknown ecological scenarios or exposure situations, which will improve the quality of ERA as well as saving time and resources. Toxicokinetic-toxicodynamic (TKTD) models for lethal effects have already proven the ability to identify patterns in effects across compounds and species. The Dynamic Energy Budget (DEB) theory may have the potential to provide a general modelling framework for sublethal effects. Models based on DEB theory have been used in ecotoxicology for decades, and these models are currently under discussion as standard approach for risk refinement at the level of tier-2. Models based on DEB theory allow for mechanistic interpretation of effects on feeding, energy expenditure, growth and reproduction. The same modelling framework can be used for all organisms, which is crucial for across - species extrapolation of effects. Many compounds, especially those that target the nervous system, act on the ability of the organisms to feed or assimilate energy. Thus,
predicting effects on feeding and assimilation is a crucial characteristic for a TKTD model to predict sublethal effects in the context of ERA. We present here the results of the i-ERA project (integrated ERA) on the responses to low food conditions / feeding impairment in four fish species (rainbow trout, fathead minnow, zebrafish, and medaka). We tested the DEB model for predicting organism level responses of juveniles (rainbow trout) and adults (all others) under low food conditions. We find that for low food conditions, fish do not change their metabolism compared to the standard DEB model. This indicates that the model can be used in ERA for the four fish species analyzed here to predict effects of compounds that act on feeding inhibition without any adaptation. The differences in the organism-level response to low food conditions / feeding impairment between the four species can be explained by differences in their model parameters. The standard DEB model can be extended to different feeding efficiency and low food conditions. We suggest extended model adaptations needed in such case, and discuss how the model can be used in risk assessments for weight-of-evidence in tier-1 and tier-2 as suggested by EFSA.

M0358 TK-TD modelling as additional line of evidence in the risk assessment for aquatic macrophytes: chlorotoluron as a case study
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To assess effects of the use of plant protection products based on chlorotoluron as active substance, various designs of laboratory tests with Lemnna sp. and other species and also mesocosm studies including different macrophyte species are available. Since it is now time to move from provability to being more predictive, this scenario, TK-TD modelling was used as an additional approach to address the potential effects of short-term exposure as predicted for some FOCUS surface water scenarios. The Lemnna TK-TD model developed by Schmitt et al. (2015) was used to simulate laboratory tests assuming exponential growth as observed in the experimental controls. Growth under field conditions was modelled as dependent on a set of factors including light conditions as well as water temperature. The substance-specific TK-TD parameters were calibrated using the results of a growth inhibition test with 7 days of exposure followed by 7 days of recovery in freshwater medium without test item. The so calibrated model was verified by comparing its predictions with results of three other tests with different exposure patterns, some of which were designed with this purpose in mind. Modelling choices were close to or above 0.9 for all four tests and, thus, the model was considered suitable for simulating effects of different exposure patterns on the growth of Lemna. We simulated laboratory refined exposure tests with PEC profiles of the 7 days worst-case time window of the FOCUS step 3 scenarios as well as field populations using the full FOCUS profiles as inputs. For the exposure profiles characterized by short-term pulses, margins of safety were above 10 to reach a 50 % inhibition of the growth rate over 7 days, the endpoint used in Tier 1. For the simulated field tests, maximum deviation of biomass under control and exposure conditions was used as assessment endpoint. If up to 25 % deviation of biomass of an exposed population from a control population is considered a negligible effect, the Margins of Safety was above 20 all analysed scenarios. The experimental result, which is variable on different conditions as well as different substances, shows that the exposure profiles considered here will, with a high probability, not lead to unacceptable effects on macrophytes. This project demonstrates the usefulness of modelling as an additional tool in risk assessment of plant protection products, particularly for extrapolation between scenarios which cannot all be tested experimentally.

M0359 TK-TD modelling as a tiered approach to reveal interspecies variability of toxicity in fish
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Species sensitivity distribution (SSD) analysis can be used in higher tier risk assessment to describe the variation in sensitivity of a group of species to a certain contaminant. Contrary to the standard procedure in which toxicity endpoints are derived by considering only effects at the end of a constant exposure experiment, this method has the potential to additionally make use of time-variable exposure and organism response over time. Here, changes in SSD (and the corresponding HC5s) can occur for the same species using the toxicokinetic/toxicodynamic (TK/TD) model for survival GUTS. The GUTS model was parameterized based on standard acute tests for each fish species and both stochastic death (GUTS-SD) and individual tolerance (GUTS-IT) assumptions were tested. Then, the 58-day LC50 for constant exposure and for two different pulse exposure scenarios (single and double pulses) were derived. The LC50 values were subsequently used as input provided the SSD calculations. The SSDs were derived by fitting probability distributions to the LC50 data, and the corresponding HCs5 were determined. The analysis was performed separately for two compounds. Results with both toxicons revealed that the sensitivity ranking for the fish species and consequently the HC5 values were not the same among the tested exposure scenarios. Predictions with either GUTS-SD or GUTS-IT models also had an impact on the SSDs and did not yield the same results for the same exposure profile. Additionally, longer exposure durations did not always result in lower HC5s. These findings infer that SSD strongly depends on the exposure scenario, and reveal the interference of substance toxicokinetics and organism responses to toxicity in determining the sensitivity ranking of the species. Therefore, it is essential for a reliable environmental risk assessment not only to consider realistic exposure scenarios, but also the TK/TD processes related to the substance and the organism. With a set of standard data, the GUTS model can help to achieve this goal for untested exposure patterns.

M0360 RIFCON EasyGUTS: Ready-to-use and freely available software for TK/TD modelling of survival
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GUTS (General Unified Threshold model of Survival) is one of the most commonly used models used for toxicokinetic and toxicodynamic evaluations of aquatic experiments in the context of the European registration of plant protection products at lower tiers in the ecotoxicological risk assessment. One user friendly implementation of this model is the Windows based program EasyGUTS. This implementation and its functionalities were recently tested and verified using published data. Results obtained with EasyGUTS are in good agreement with results obtained from various other publications and model implementations. However, one limitation of the program during this verification process was that it was only possible to select the log-normal distribution for the individual tolerance model rather than giving the possibility to also use other functions like a log-logistic distribution. This was the case since the GUTS R package to which EasyGUTS is linked, is restricted to only this possibility. Since a TK/TD draft guideline is expected earliest next year and no preference for a distribution is given in recent publications, in line with the EFSA ‘Scientific Opinion on Good Modeling Practice’. Moreover, EasyGUTS as a functional tool was tested in internal and external modelling workshops. Our experience is that the usability of the software and the robustness of the calibration algorithm was fitting well, so that even all users could reproduce results and decisions. Since EasyGUTS is finally verified and harmonised with the R GUTS package, it is ready to use under free license agreement and can be downloaded from the RIFCON homepage beginning of 2018. This poster presents the functionality of EasyGUTS and gives insight on the sensitivity of the model to initial parameter values and the influence of different distributions used for the individual tolerance model.

M0361 A new test design to inform TKTD models on species sensitivity
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Recently, several TK/TD population modelling approaches have been developed and are applied in different risk assessment areas. The European Food Safety Authority (EFSA) guidance on tiered risk assessment for plant protection products for aquatic organisms in edge-of-field surface waters states GUTS modelling as an approach for the aquatic risk assessment for the evaluation of time variable exposure. For aquatic invertebrates two key questions exists for the ecological threshold option. 1) What is the species sensitivity distribution 2) What are the effects from short-term / peak exposure patterns on long-term survival and reproduction. Currently TK/TD models are parameterised on the standard Tier 1 or Tier 2 scenarios. Particularly in chronic tests, toxicodynamic effects are exposed over long time periods (week to months), which makes these experiments costly, time consuming and which limits the number of non-standard species to be investigated, as these species - in the absence of appropriate husbandry and test methods and due to complex biology - are particularly challenging to test reproducibly in chronic set-up's. Here we will present an approach using the GUTS model (TK/TD model for survival) informed by specifically designed peak-exposure experiments to answer both questions. We will employ short-term experiments, lasting 48h with two short peaks of 4h duration, at 3 different treatment levels of an insecticide, in combination with several observation time points for 5 aquatic insect species and 3 crustaceans. The outcome of these experiments will inform the GUTS model and will allow for a suitable calibration, after which it will then be possible to construct pattern-specific species sensitivity distributions to be used in acute effect assessments for time variable exposure patterns like FOCUS scenarios. Moreover, the approach may also provide further insights whether peak exposure experiments in an acute study design can be used to investigate sensitivity differences also on a longer-term time scale, by concurrently preventing the pitfalls and potential artefacts arising from not yet developed, adequate long-term husbandry- and test protocols for non-standard species.

M0362 Impact of temperature on species sensitivity distribution in aquatic invertebrates
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Aquatic effect assessment uses results from laboratory experiments at constant environmental conditions. However, for these tests organisms are kept under optimal temperatures which might differ across species. If done for multiple species, results from these toxicity tests are used to statistically derive community level endpoints, such as the HC5, from species sensitivity distributions (SSDs). Therefore, data from acute toxicity tests are ranked using cumulative distribution. Apparatus toxicity overviews such as the LC50s, have been reported to depend on ambient temperatures and particularly in aquatic invertebrates and fish and if comparing species sensitivity the results might be biased by the experimental conditions. It has been demonstrated that changes in physiological rates with different temperature regimes can be described by the Arrhenius function. Part of this study is to examine if the Arrhenius function is also able to predict TKTD model rates, such as GUTS, for different temperatures. We use GUTS to extrapolate toxic effects across temperatures and investigate the impact of temperature on a species sensitivity distribution (SSD) with aquatic invertebrates regarding Chlorpyrifos.

MO365 The use of population models in copper risk assessment: a case study with Aciensper transmontanus
K. Vlaeminck, Arche consulting / GhEnToxLab; K. Vlaene, Ghent University / GhEnToxLab; P. Van Sprang, ARCHE; K. De Schamphelaere, Ghent University (UGent) / Applied Ecology and Environmental Biology
Current metal risk assessment consists of assessing single-species data on metal toxicokinetics and constructing species sensitivity distribution (SSD) for the derivation of safe thresholds. Despite their usefulness, SSDs have been criticized over the last decades for being ecologically unrealistic, and for typically only accounting for individual-level endpoints. Population models as an alternative are becoming more popular in ecotoxicology as they translate a pollutant’s effects on individuals (e.g. survival) to the population level (e.g. growth rate). Additionally, ecological models are less expensive and time-consuming to develop and perform research with compared to population experiments. In this study, we aimed at adapting an existing white sturgeon (Aciensper transmontanus) population model to predict population level effects of copper toxicity. The white sturgeon is a fish species particularly sensitive to copper during early developmental life stages. An individual-based model (IBM) was implemented using the software platform NetLogo. Copper effects were integrated by adjusting the mortality rate for the sensitive life stage (age 0 individuals) For different scenarios (i.e. environmental configurations, exposure profiles, etc.) population-level effects were assessed as a function of the copper concentration. As expected, population equilibrium density decreased with increasing copper concentrations. Effect concentrations (ECx) values for population equilibrium density were situated in the same range (as traditional lethal concentrations (LCx) values) at the individual level. Nonetheless, the magnitude of the population’s response to copper depends on several environmental factors such as habitat fragmentation and distribution of the pollution in the river system (random, heterogeneous, or homogeneous). Population ECx values were derived with the IBM by extrapolating observed (conventional) LCx values from literature. However, the adapted population model for A. transmontanus contains some inherent assumptions which need further fine-tuning. By investigating the mortality profile (i.e. mortality over time) in depth, the mortality sub-model could be improved further, increasing predictability of the model. Additionally, investigating population density-dependent effects on the survival of age 0 individuals could increase accuracy as well. This study shows that population models could be used as more ecologically-relevant tools in metal risk assessment.

MO366 Comparison of toxic effects on Daphnia magna between a metal, a pesticide, and a PAH, in a toxicokinetic-toxidynamic framework
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Modelling techniques are becoming more prominent in the risk assessment of chemicals. Mechanistic models, such as individual-based models (IBM) with a foundation in the dynamic energy budget (DEB) theory, are increasingly promoted as alternative tools in ecological risk assessment. In this context, lethal toxicokinetic-toxidynamic (TKTD) models are often used to describe sub-lethal effects on the life cycle of the modelled organism. Inherently, the mode of action will differ between compounds (i.e. compounds will affect different physiological processes). The current study compares TKTD parameters of three different compounds, and examines their influence on the dynamic energy budget (DEB). The DEB model (Daphnia magna) is made up of three different heat components: a heavy metal (Cu), a pesticide (endosulfan), and a poly-aromatic hydrocarbon (pyrene). The TKTD model was calibrated for each compound based on life cycle experiments with Daphnia magna effects of the three compounds. During life cycle experiments (21-days), growth, reproduction and survival were monitored at different concentrations for each of the compounds. Using all three endpoints, the modes of actions and the TKTD parameters were estimated for copper, endosulfan, and pyrene. Combining the TKTD model with DEB-IBM, effects on physiological processes can be translated to the organism level.

MO367 Deriving predicted no-effect concentrations for perfluoroalkyl acids in the Po river ecosystem through a novel methodology based on the AQUATOX ecosystem model
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Health and environmental risks posed by perfluoroalkyl acids (PFAs) have been the establishment of safe ecological thresholds such as Predicted No-Effect Concentration (PNEC), based on procedures incorporated in the REACH regulation and Water Framework Directive in and related guidelines. These policies offer three methodologies for deriving PNEC: use of assessment factors (AF), species...
sensitivity distribution (SSD), and results from model ecosystems and field studies whose task is to extrapolate single-species data to ecosystem-level responses. Although AF and SSD methods are described by strict guidelines making them commonly applied, they do not consider the effects of ecological interactions between species on the assessed risk level, which is potentially not-negligible since population dynamics in polluted environment are not only driven by direct toxicity of chemicals on single species. One cost-effective alternative for assessing ecological risk of chemicals considering also indirect ecological effects is the use of mechanistic ecosystem models, simulating the multiple interactions between biotic and abiotic ecosystem compartments. However, there is lack of official guidance for models choice, development and use, resulting in scarce implementation of ecological models for regulatory purposes. Accordingly, two main goals of this work were to: (i) to derive a method for deriving PNEC by use of the US-EP AQUATOX ecosystem model, and to evaluate the risk posed by PFAAs (represented by two long-chained and two short-chained compounds) in the ecosystem of the Po, the greatest river in the Northern Italy. Through AQUATOX, water concentrations of PFAAs resulting in a non-negligible biomass loss for each modelled population of the ecosystem were assessed, thus connecting biomass density (a model output) to a “safe” concentration (PNEC). The resulting PNECs were compared to PNECs derived with conventionally used AF and SSD methods to assess the performance of the proposed novel methodology.

MO368
Incorporating spatially explicit metapopulation models as the endpoint of an Adverse Outcome Pathway-based Bayesian Network-Relative Risk Model
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Population viability analysis is useful tool for assessing the environmental risk of toxicants because it produces endpoints relevant to managers and can be manipulated to compare the potential outcomes of conservation actions. In general, many Environmental Risk Assessments (ERAs) lack utility and realism because they fail to incorporate the combined effects of lethal and multiple sub-lethal impacts, environmental stressors, and chemical mixtures into a relevant endpoint for managers. To improve the utility of regional scale risk assessment, we are developing a Bayesian Network-Relative Risk Model that incorporates the combined effects of toxicants and environmental stressors into an Adverse Outcome Pathway (AOP) framework linking environmental conditions to spatially explicit metapopulation models. As a primary case study for this new model, we are examining the impacts of organophosphate (OP) insecticides on ESA-listed chinook (Oncorhynchus tshawytscha) and coho (Oncorhynchus kisutch) salmon populations using site specific data from the Lower Skagit, Nooksack, Cedar, and Yakima River watersheds in Washington State. The AOP within the BN-RRM links concentrations of OPs to % Acetylcholinesterase (AChE) inhibition which is then linked to sublethal impacts that are incorporated into matrix metapopulation models through age-specific reductions in survival and reproduction. The outcome of this effort will be an adaptable management tool that uses existing, disparate data to link realistic toxicant concentrations to probabilistic population outcomes. The preliminary results of this model development suggest that environmentally realistic concentrations of OPs may slow the growth of salmon populations, undermining the success of current restoration efforts.

MO369
Modeling and monitoring the effects on the central nervous system of a chronic exposure to low dose of pollutants: an innovative strategy with first results
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Chronic low dose exposure and possible cumulative effects of various pollutants could affect consumer health and may contribute to the development of neurodegenerative pathologies. Due to the highly complex and long exposure, a clear association between exposure and disease has often been suspected, but rarely proven. An additional drawback is the high diversity of in vitro models (cancer cell lines, stem cells, primary embryonic cells), bringing additional complexity in the deciphering of the observed effects of pollutants. To properly assess the risks and to reevaluate the maximal acceptable dose of specific pollutants in the food chain, there is a need for efficient modeling of pollutant effects on the central nervous system (CNS). To address this need, we are developing a new approach to evaluate the consequences on neuronal health of long time exposure to pollutants. We are actually re-evaluating the neurotoxic effects of chlordecone (CLD) as proof-of-concept of our strategy. Several concentrations of CLD were used to treat a variety of mouse primaries isolated from different postnatal CNS areas. We then assessed neuronal functions using specific markers for neuronal death, neurite development and synapse formation. In parallel, we produced cerebrospinal fluid (CSF) from pigs exposed to CLD via contaminated food. This CSF containing CLD and its by-products that are able to cross the blood brain barrier could then be used on the same cultures to compare its effect with that of porcine CLD. Following direct treatment with the molecule. We were able to identify a direct neurotoxic effect (10 µM) on specific purified neuronal cultures together with more subtle damages at lower concentrations including neurite arborization defects (0.1 to 1µM depending on the respective neuronal cultures). Additional effects on pure glia cell cultures at higher concentrations (100 µM) suggest a reactive gliosis in the whole animal. These observations were confirmed by CSF treatment using CLD-contaminated CSF but not with CSF from control pigs. In addition, the dose with no observable effects is at least 10 times lower using primary postnatal neuronal cultures compared to embryonic cultures. Our experimental model is therefore much more sensitive and may reflect more precisely the consequences of chronic CLD exposure. Our strategy could help to re-evaluate the CNS effects of this remnant pollutant present in West Indies soils.

MO370
A new classification method for mechanisms of toxic action
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A knowledge of the mechanism of action (MechA) of substances is a crucial first step in risk assessment approaches, especially when using in silico models to predict (eco)toxicity. Mechanisms of Action are similar to Molecular initiating Events which govern molecular interactions between xenobiotics and biological material. Using the accumulated knowledge of MechAos covering hundreds of molecules, we developed a set of structural alerts associated with specific MechAos. Consequently, a new method to predict MechAos with high accuracy and with simple rules was developed (Bauer et al 2018). We compared this method with Verhaar (as updated in 2008) (Verhaar et al., 1992, 2000, Enoch et al., 2008) and Russom (MOA classification by OASIS implemented in OECD QSAR Toolbox) (Russom et al., 1997) methods, and our decision tree showed the best statistics. This method is currently being implemented into a software, and it will be made freely available and we consider it as a useful support in risk assessment. This model will be especially enhanced with the addition of new rules and minor corrections as needed.

Biocides and Veterinary Medicines: latest developments in regulatory risk assessment, research and monitoring (P)

MO371
Biocide leaching from building facades: Pseudo-persistence in soil due to reoccurring emissions
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Facade paints and render are commonly protected against biological deterioration using biocides. Mixtures of in-can as well as film preserving bactericides, algaecides and fungicides are added to the materials. Nevertheless, active ingredients leach from the treated facades, if contacted with wind-driven rain. Especially in suburban residential areas a large fraction drains directly to soil, e.g., flowerbeds, gravel strips or the lawns surrounding the houses. Consequently, the soil in areas with biocide-treated buildings is exposed to rain runoff water highly polluted with biocides. In the present study, the degradation rates of eleven biocides in soil were determined in laboratory microcosms. Degradation half-lives ranged from rapidly degrading (T½ < 10 d) to compounds with higher persistence (T½ > 120 d). For two selected biocides (terbutryn and octlisothiazoline) a set of transformation products were quantified in the microcosms as well. This showed that the mass balance for terbutryn could be closed with nine analysed transformation products for the entire incubation period (120 d), revealing that relative persistent metabolites are formed. In contrary, the mass balance including transformation products for the entire period was not closed, as the number of transformation products were quantified. Reduced toxicity of transformation products towards Alivibrio fischeri than the...
regardless of the type of rodenticides used. This analysis showed that the main loss mechanisms are the transport of biocidal residues to the environment and the leaching of biocidal active substances during the polymerisation and curing process of the façade coatings. The leaching rate of biocidal active substances into the environment depends on the nature of the pigments used, with some pigments being more effective at slowing down the leaching rate than others. For example, in one study, it was found that pigments with a high metal content were more effective at reducing the leaching rate of biocidal active substances than pigments with a low metal content. This is important for the risk assessment of biocidal products, as it highlights the need for further research into the effects of different pigments on the leaching rate of biocidal residues into the environment.
A case study on exposure assessment of biocides in PPCP using exposure assessment models

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Several accidents caused by the use of chemical products created a need for risk assessment of chemicals used in Pharmaceuticals and Personal Care Product (PPCP) in Korea. CMIT/MIT which is a mixture of 5-chloro-2-methyl-2H-isothiazol-3-one and 2-methyl-2H-isothiazol-3-one is used in PPCPs as preservative and biocidal agent. Despite of its inhalation toxicity, this chemical has been used as humidifier contaminated from the 1990s without considering its exposure route and caused a lot of victims to suffer from its adverse effect. In contrast, CMIT/MIT was detected in toothpaste and the products containing the mixture were recalled in 2016 even though its toxicity via oral route is not known. The aim of study is assessing the exposures of CMIT/MIT in PPCP and comparing derived from these two different model was identified. It is concluded that difference between exposure calculation equations and input values affects the results. And each level of model has its strengths and weaknesses. Several improvements are needed to apply European models assessing in reflection of Korean exposure scenarios.

Hazard evaluation of biocides and its metabolites for the aquatic compartment
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The LIFECOMBASE project main target is to promote the sustainable use of biocidal active substances by developing prediction models. As a first step, a database compiling aquatic toxicity data to the aquatic compartment for biocides and derived metabolites has been implemented. The aim of the present study is to make a critical review of this information in order to have a wide view about their potential risk for the aquatic compartment. All the biocides approved and under review were compiled (277 active substances; Regulation (EU) 528/2012). A selection of 192 biocides with possibilities to be modeled and a search using several official and scientific databases, looking for any possible metabolite derived from their release in the aquatic compartment were done. Data was collected in an excel file, including identification of the biocide or metabolite (EC NR, CAS NR, SMILES), classification data (main group, product type, regulatory status), LogP, hazard assessment and exposure scenarios. Based on the found data was related to toxicity in fish, followed by invertebrates and algae, marine microorganisms being the least studied. There was not reported data for around 80% of the 185 metabolites found, probably due to, in some cases, their commercial unavailability. Another identified problem was that some data were developed with formulated products or with active substances for which purity was not reported. Data already analyzed for the acute toxicity indicated that, 62% of the biocides were located in category 1 for invertebrates, 54 % for fish and 52% for algae. Only 25% biocides belong to this category for the microorganisms group. Metabolites are mainly less toxic than the parent biocides, however many of them present the same toxicity and very few (< 7%) are more toxic. The ongoing work indicates that biocides and a considerable percentage of their metabolites present a high toxicity for the aquatic species. It also identifies data gaps related to the ecotoxicological potential for metabolites.

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Synchronous decreasing levels of imposex and tributyltin (TBT) in dogwhelk (Nucella lapillus) from Norway, 1991-2015

Imposex is TBT-induced permanent male sex-character in female dogwhelk (Nucella lapillus). This biological effect is quantified by the Vas Deferens Sequence Index (VDSI). Levels of imposex and TBT in N. lapillus have been monitored annually in Norway since 1991. Populations around the North Sea were critical reduced in the 1990s, due to increased use of TBT-based antifouling paints. Before the global TBT-ban in 2008, increased TBT-levels coincided with increased imposex prevalence at many monitoring sites located close to high maritime activity. After 2008, decreasing TBT-trends at former impacted sites, lead to population recovery of N. lapillus. The observations in N. lapillus further corroborated by monitoring data showing decreased TBT levels in blue mussel (Mytilus spp.). This monitoring data confirms the rationale of implementing strict ambient regulations on industrial chemicals when these can be linked to ecological perturbations in coastal ecosystems. The TBT/imposex monitoring was conducted at eight coastal stations representing the Norwegian coast from the Oslofjord to the Varangerfjord, following the guidelines given by OSPAR and ICES. Subsequently, 50 specimens from each station was analysed individually for imposex/VDSI and pooled (only females) for TBT and other organotin like triphenyltin (TPTTN). TBT and VDSI indices (VDSI= 0.828 µg/kg w.w. and TPTTN= 0.3 µg/kg w.w.) were low in N. lapillus at eight stations in 2015. At most stations, VDSI was 0 or close to 0 and below the OSPARs Background Assessment Criteria (BAC=0.3). The highest level (VDSI= 0.828) was found at the shipping channel Karmusundet, which were above BAC but below the OSPARs Ecotoxicological Assessment Criteria (EAC=2). There were significant downward long-term (whole period 1991-2015) and short-term (recent 10 years 2006-2015) trends for both imposex/VDSI and TBT based on time trend analysis. These results show that the Norwegian legislation banning use of TBT on boats less than 25 m in 1990, on larger ships internationally from 2003, and the total ban in 2008 have been effective.
in reducing imposex in N. lapillus and have re-established some of the populations. Low levels or significant downward long-term and short-term trends for TBT in common periwinkle (Littorina littorea) and blue mussel (Mytilus spp.) substantiate this.

Risk assessment issues for algacides under BPR

A prospective and comprehensive environmental risk assessment (ERA) must be performed on the active substance for the market authorisation of biocidal products. The technical guidelines are made publicly available by ECHA. The data required for the ERA include the determination of a set of properties (physical-chemical, fate parameters, short- and long-term ecotoxicity), an effective in-use dose or concentration, frequency of application of the biocidal product etc. . . . In coming years, a prospective risk assessment has to be prepared in order to address the risk associated with the use of biocidal containing these active substances and also any substance of concern. The initial approach for a prospective environmental risk assessment will proceed with a Tier 1 calculation, which assumes 100% of the applied chemical will be released and ignores the formation of degradation products neither biodegradation of the active substance, neither consumption of active substance due to its biocidal activity. These initial assumptions may lead to an overestimation of the environmental exposure and risks to the active substance. The initial environmental risk assessment can be refined with supporting data e.g. on the degradation and/or dissipation of the active substance and also by consideration of risk management measures to reduce exposure. In the case of biocidal products applied in swimming pools to disinfact or to control algae growth in water, several actives substances are under evaluation or are recently approved in the EU which includes halogenated compounds, inorganic compounds, quaternary ammonium compounds. The present work focuses on the application of the Biocide risk assessment methodology to algacide applied in swimming pools. The post work is focused on following key steps: (i) define an effective in-use concentration which is an input parameter for assessing the risks associated with the active substance to define the ecotoxicological dataset which is needed in order to determine accurate PNEC values for characterising the risks posed by the active substance to discuss possible options to refine the exposure of environment including new studies and risk management measures

Could a spatially distributed modelling approach enhance post approval considerations for veterinary medicines?

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Tiered chemical risk assessment framework adopted in Europe for assessing the surface and groundwater risk from veterinary medicines used to treat livestock follows a tiered approach. The initial exposure assessment is a simplistic approach, with the FOCUS suite of models (FOrum for Co-ordination of pesticide fate models and their USE) often subsequently required for higher tier surface and groundwater refinement. Standard FOCUS scenarios defined within the guidance are intended to represent worst-case scenarios, with results being used to assess the need to consider additional fate and effects which are not adequately represented within the standard scenarios. This tiered approach to calculation of PECsoil and PECgw involves using a tiered approach to calculate PECsoil and PECgw of VMPs from livestock use dose or exposure, namely in surface and groundwater. However, this tiered approach may not account for some of the complexities of real-world exposure scenarios, such as the influence of environmental factors on the fate and transport of veterinary medicines in the environment.

Veterinary medicine products (VMPs) are used in livestock production to preserve animal health or to promote growth in certain categories of animal; feed additives (FAs) are products aimed at improving the quality of feed and the quality of food from animal origin, or to improve the animals’ performance and health. These substances may not be put on the market unless authorisation has been given following a scientific evaluation demonstrating that they have no harmful effects, on human and animal health and on the environment. In particular, according to European Framework Directive 2001/82/EC, the environmental risk assessment (ERA) procedures for VMPs are based on technical guidance documents which propose a tiered approach to calculate PECsoil and PECgw of VMPs from livestock manure spread on the field. On the same way, the ERA procedure for feed additives is reported in a technical guidance document from EFSA which describe a two-tiered approach to calculate PECsoil and PECgw from spread manure.

Calculation of PECsoil proposed by the two ERAs in the first tier is directly related to the “annual nitrogen (N) immission standard” which is the amount of nitrogen per Hectare spread on or into the field. Both ERAs propose a default value of 170 kgN Ha⁻¹ which is the maximum allowed annual amount of nitrogen originating from animal manure on a farm within nitrate vulnerable zones (NVZ). On the other side, in Europe, NVZs are measured, and in total 25% of substances actually distributed on these zones higher thresholds of N immission standard are allowed. Both ERAs procedures could therefore underestimate the PECsoil with a potential environmental toxicity for non-target terrestrial organisms. This study is aimed to evaluate if PECsoil, calculated using standard models currently used in the authorisation procedures of VMPs and FAs, are sufficiently adequate to protect soil and groundwater. In the case of FAs applied in swimming pools, for instance, the exposure is characterised by low application rates, repeatedly repeated, and long exposure in surface water. In the case of VMPs applied in livestock, the exposure is characterised by high application rates, rarely repeated but occurring on a larger scale. In this case, the exposure may be characterized by high surface water concentrations due to the larger application area, which may lead to overestimation of environmental exposure. However, in this case study, we considered Lombardia Region in Italy which shows one of highest livestock manure loads in Europe. Information on manure use at field scale were gathered from Valoréf system, which is currently used in Lombardia Region to improve the manure management.

Quick scan to monitoring data of veterinary pharmaceuticals in the Netherlands

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On the Dutch market, approx. 260 active substances are used in different veterinary medicines. In a quick scan, we investigated the potential contribution of veterinary medicines to environmental exposure of animals to environmental concerns of veterinary medicines in Dutch waters to feed possible policy measures. For this, we gathered information from detected compounds in both groundwater and surface water, as these sources are related to drinking water production. From the bulk of the compounds on the market no measurement data are known (84%). Of the 260 compounds used in veterinary medicines, only 82 are measured, and in total 25% of substances actually distributed on a measurement basis. Detection involved 15 antibiotics, four anti-parasitic resources, three anthelmintics and one painkiller. Our quick-scan confirms that a good insight into the presence of veterinary medicines in the water cycle is still lacking. Not only measurement data is limited, it became clear that data on local and regional surface waters is missing or fragmented. We propose inclusion of monitoring data in national and international databases, so that becomes available for other purposes, including prioritization. We also see that current monitoring schemes is not specifically aimed at veterinary medicines, and due to the fact that emissions, temporal and spatial trends, emission routes and concentrations at relevant locations are little available, no clear overview of risks is yet existing. We further noted that the origin of a detected compound cannot always be properly traced back to the primary use for only. We note that veterinary compounds are also used in human medicines or as pesticides. Admission to the market, based on active substances, is therefore sometimes regulated in different rule-making and also, usage data is scattered. This makes priority setting difficult when performed in a segregated view on the universe of chemicals.

Comparing methods for estimating environmental emissions


The environmental risk assessments consist of information on exposure and hazards of chemicals to environmental compartments. Environmental emissions for biocidal products are estimated according to Emission Scenario Documents (e.g. OECD ). In some ESDs, the emission is calculated based on the use/consumption of the product as specified on the label (e.g. PT18), with a number of default assumptions applied. However, other ESDs include the facility to take account of tonnage information as well as average consumption values in product specific calculation models (e.g. PT2). Both approaches have strengths and weaknesses. However, the importance of establishing realistic and reliable methods of estimating environmental emissions cannot be understated, especially in light of the intention to develop guidance on aggregate assessment. Accordingly, this paper will illustrate on two approaches for estimating emissions: regional tonnage (top down) or consumption (bottom up). Opportunities and limitations of the applicability of the data and their implications for use in EU environmental exposure assessment will be evaluated.
Interpretation and uncertainty - overcoming challenges of translating LCA results into reliable information (P)

MO387

Recommendation on Steam Cracker allocation for the sake of comparability of petrochemical products datasets used in LCA studies

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The steam cracker process turns fossil hydrocarbon feedstocks into several different main products, like ethylene, propylene, butadiene, olefins, aromatics, and so on. They are all basic building blocks of many chemicals and polymers used in nearly all products and sectors. Thus LCA data of steam cracker products directly influence a huge amount of further downstream products. It is therefore important that LCA data for steam cracker products are modelled consistently, enabling a reduction of uncertainty and a better interpretation by LCA experts, particularly in perspective of comparability, in LCA studies of these downstream products. Basing on ISO 14044 and on the abundant existing literature on this topic the Life Cycle Thinking and Sustainability working group of PlasticsEurope, composed of experts from its member companies, plus some experts of the Chemical Sector of the World Business Council for Sustainable Development, and some LCA consultants have issued a recommendation built through a consensus 5 years long process. The presentation will elaborate on the discussions and on the recommendation finally issued, considered as the best compromise between comparability and specific representativeness. For multi-output processes, such as a steam cracker, ISO 14040 and 14044 standards define a hierarchy of several options. Due to the nature of steam cracker processes allocation is considered as the preferred option. The concept of defining a main “products” fixed list in combination with a mass-based allocation for steam crackers has led to a consistent LCA approach, independent from market prices, technological changes or market driven adaptations of steam cracker outputs. It gives practitioners a clear guidance for the allocation process. This results in less differing environmental data for steam cracker products and will lead to a higher comparability. The results are much more stable, although the same products will have slightly different LCI results depending on different data sets, as different products from the derived steam cracker. Such a collaborative work towards streamlining should be engaged for all chemicals, and should be applied within all database both to background and foreground parts, like for example in the European EF compliant database.

MO388

Actual versus default uncertainty in ecoinvent database

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Variability of national life cycle inventory flows is a relevant uncertainty source and should be properly informed in public databases. Within the scope of the Sustainable Recycling Industries project, life cycle inventories for Brazilian construction products were developed and submitted to ecoinvent following its guidelines, including the preferred use of the lognormal distribution for uncertainty modelling, which requires converting sample average and variation into the geometric mean and the unbiased variance of the underlying normal distribution. However, dataset reviewers inform that most data providers do not perform these conversions and simply use the sample average for flow amounts, frequently associated to default basic uncertainty factors suggested by ecoinvent. This work discusses the implications of three different uncertainty modelling approaches: 1) using both converted mean and variance, 2) using the sample average with the converted variance; 3) using the sample average and default basic uncertainty variance (probably the most common approach). Primary data collected in 25 concrete block factories were used in the analyses. Influence on life cycle impact assessment results was assessed using Monte Carlo simulation with 10,000 iterations, CML 1 A-h method and ecoinvent v.3.2. Results show that the sample weighted average and the geometric mean differed significantly. Therefore, using the sample weighted average as a proxy for the lognormal geometric mean may overestimate impacts, in our case by approximately 10%, considering only the effects of the concrete block production process flows. Since existing datasets may have followed this approach, the impact effect and uncertainty values are significant lower than measured variations across manufacturing sites, which is inconsistent with a conservative estimation approach. Thus, uncertainty information provided by ecoinvent might contain inconsistencies and lead to errors in uncertainty assessment, such as impact overestimation. Uncertainty modelling can be improved in the database by allowing the input of different amount parameters, performing automatic conversions in the submission software or simplifying the provision of uncertainty data using simpler probability distributions.

MO389

Life cycle assessment of battery systems with harmonized life cycle inventories considering different storage applications

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The penetration of renewable electricity has greatly increased in the past decade. Battery is a key storage technology to balance supply and demand and to facilitate the world’s transition towards a sustainable energy system. However, having a comprehensive overview of batteries’ life cycle environmental performance still remains a challenge, because battery technologies are of various kinds and the applications of batteries vary. These applications are different from each other in terms of required power and energy size as well as number of cycles. Due to these different requirements by applications, the same battery technology needs to be operated differently and sized accordingly. Numerous studies in the past investigated the life cycle environmental performance of batteries; however, most of them are focused on the application of batteries in electric vehicles, considering a limited number of lithium-ion battery technologies, while the stationary applications of batteries were less explored in limited studies. In addition, these studies are mostly conducted based on diversified sources of life cycle inventory data, without harmonizing the assumptions that are not necessarily different. Peters et al. have recently applied in real scale, the application of nanomaterials as adsorbents but they are compared without considering the applications. Another study by Baumann et al. considers the applications of battery in the assessment, without addressing the country of application, which results in partial understanding of contributions in the life cycle emissions. This study therefore addresses these challenges, by considering six battery technologies for five storage applications in three representative applications of Europe. One of the objectives of studies is the harmonization of inventory data is carried out to a greater extent. We also extend the scope of the system, which is often limited to battery pack, to include the complete balance of systems, which ensures the operation required by the applications.

MO390

LCA of nano-adsorbents - Interpretation of laboratory results

A. Kazemi, Tarbiat Modares University / Department of Environmental Science, Faculty of Natural Resources; S.I. Olsen, Technical University of Denmark / DTU Management Engineering. Division for Quantitative Sustainability Assessment; N. Bahramipour, Tarbiat Modares University / Department of Environmental Science, Faculty of Natural Resources; A. Heydari, Tarbiat Modares Nano-adsorbents as an emerging product and a special application of nanomaterials can increasingly play an important role in the control and removal of environmental pollutants. An example of this is the use of nano-iron to remediate contaminated groundwater. However, even though particularly this example has been recently applied in real scale, the application of nanomaterials as adsorbents is still an emerging technology at the early stages of development. Hence, this study enables an environmental assessment of nano-adsorbents as an emerging product/technology based on the results from the laboratory. Two nano-adsorbents with graphene-based (MGO-NH-SH) and FeO3-based (FeO3@SiO-NH-SH) composites, which function with a similar thiol group for Hg(II) removal are comparatively different stages of the production. Removal of mercury is important due to its historic cases of fatal contamination and its continued use. Although mercury must be removed from the contaminated sites it is still very relevant to make an LCA in order to ensure a balance between the impacts of producing the nanoadsorbent versus the avoided impact of the mercury that is being removed. The environmental impacts of synthesised adsorbents including energy use, climate change, water use, human toxicity, and ecotoxicity are investigated by a stepwise procedure during their synthesis processes, regarding their potential to remove mercury from polluted water (functional unit is removal of 1 kg of Hg(II)). Accordingly, characterization results showed that although the process of the functionalization of nanoadsorbents leads to the increase of the adsorption capacity of them, it is also associated with a significant enhancement of negative environmental impacts. A “what-if” perspective was applied to assess the uncertainties of using lab-scale data for parameters including amounts of acid (HCl + H2SO4), ammonia, ethane, methanol, DCC (N,N'-dicyclohexylcarbodiimide), NHS (N-Hydroxysuccinimide), water recovery, and electricity. The results of test comparing the impacts between MGO-NH-SH and FeO3@SiO-NH-SH estimated comparatively 37, 34, 40, 31, and 26% more climate change, water use, human toxicity, and ecotoxicity, respectively for the latter. Sensitivity analysis was employed to determine the uncertainties for scale-up production and it is shown that especially potential reductions of electricity use, ethane and DCC can reduce the impacts significantly.

MO391

Quantifying the influence of consumer behaviour on water, energy and greenhouse gas footprints of showering

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Life Cycle Assessment (LCA) has been used as a tool for environmental footprinting of a wide range of household cleaning activities. Even though differences in the way household activities are performed by consumers may alter the outcome of LCAs, the variability in consumer behaviour is generally ignored in LCAs, which use the average behaviour as a basis for generating the environmental impacts. The goal of our study was to demonstrate how the data on consumers’ reasoned choices, consumers’ habits, climatic parameters, manufacturing of products and infrastructure of countries can be combined to quantify the variability in the energy use, greenhouse gas emissions and water footprints related to the life cycle of showering. Data for showering were modelled in 4 countries namely Australia, Switzerland, the United Kingdom and the United States using various data sources to quantify the associated variability. Results showed that both inter-country behavioural, climatic and infrastructural differences as well as intra-country variation in consumer behaviour are crucial for determining the variability in the life cycle environmental impacts. Inter-country variability – the ratio between the highest median footprint and the lowest median footprint over the four countries - in the 4 main output variables of the model i.e. energy use, GHG emissions, water withdrawal, water consumption and water scarcity was a factor of 1.5, 2.2, 1.4 and 5.8 respectively. Intra-country variability - the ratio between the 25th percentile and the 75th percentile of the distribution - was typically higher than inter-country variability and ranged between factors of 5 and a factor of 20 depending on the process and indicator considered. Sensitivity analysis showed that consumers’ reasoned choices - particularly heater type and shower flow rate- and their habitual behaviours - particularly shower duration-, are the dominant sources of variabilities. Reductions in the water and energy related impacts of showering through changing of reasoned choices are achievable by one-off decisions such as buying an energy efficient water heater. However, reducing the impacts through changing of consumers’ habits could be challenging and needs more systematic approaches as consumers tend to keep their old habits.

MO394 Ecotoxicity and fate of Ag and CeO2 nanomaterials in outdoor lysimeter experiments

Nanomaterials (NM) will enter the environment via diverse pathways. Sewage sludge for example is repeatedly applied as fertilizer on farmland due to its high nutrient content. This may lead to a significant increase in the occurrence of NM, particularly heater type and shower flow rate due to returning digestate from biogas plants to the field used for energy production. This project strives to develop robust but also generalisable statements about nitrous oxide emissions and ammonia volatilisation due to returning digestate from biogas plants to the field used for state-of-the-art agricultural methods. The results will support the environmental assessment of WOSR production including additional aspects such as fertilizer use efficiency.

The environment as a reactor determining fate and toxicity of nanomaterials (P)

MO395 Site-specific N-emissions of rapeseed cultivation in Germany
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Globally growing population increases the demand for food, which should be produced as efficiently as possible but also environmentally friendly as possible. Simultaneously climate change requires the reduction of greenhouse gases (GHG) in order to keep the global temperature increase below 2°C. Germany has defined ambitious goals to reduce its GHG-emissions. The reduction targets are 40% by 2020, 55% by 2030, 70% by 2040 and 80-95% by 2050. Within the last 15 years, GHG emissions from agriculture have not decreased. Simultaneously the European Water Framework Directive requires a good status of water bodies, which is in particular regions in Germany not achieved. Winter oilseed rape (Brassica napus L., WOSR) is the major oil crop cultivated in Germany. Nitrogen field emissions are usually estimated using IPCC-emission factors that are not specific for the crop and associated with strong uncertainty. N2O field emissions are controlled by N fertilization and dominate the GHG balance of WOSR cropping due to the high global warming potential of N2O. The same applies for nitrate emissions that dominate the Eutrophication potential or ammonia emissions for the Acidification potential of WOSP when organic nitrogen fertiliser is applied. To address these issues and support decision makers, our project aims to educate specific emissions factors for WOSR and that can be included in the regional life cycle assessment studies for WOSR cultivation. Thus, field experiments were conducted to increase the data basis and subsequently derive WOSP-specific emission factors. Furthermore, the project strives to develop robust but also generalisable statements about nitrous oxide emissions and ammonia volatilisation due to returning digestate from biogas plants to the field used for state-of-the-art agricultural methods. The results will support the environmental assessment of WOSR production including additional aspects such as fertilizer use efficiency.

MO396 Recommendations on the Creation, Management and Use of Data Quality Information for Life Cycle Assessment
A. Edelean, ORISE; W. Ingwersen, US EPA

The average life cycle assessment (LCA) model can combine hundreds or even thousands of data points in order to describe a product system. LCA practitioners and generators are very familiar with the labour and time intensity, which accompanies data collection and processing. While the amount of life cycle inventory (LCI) data is continuously growing, and there are efforts to improve access to these data, questions of ‘best fit’ data and the appropriate use of results in supporting decision making still plague the LCA community. Ultimately, these are questions of data quality. Updates to the Weidema 2013 pedigree matrix include flow and process level data quality indicators. Five indicators resembling those of Weidema et al. (Weidema et al. 2013) matrix are described at the flow level. Two new indicators are presented at the process level. In previous works, all these analysis indicators were not necessarily orthogonal, in that the indicators were capturing overlapping information. In the updated table, all indicators are independent. The adaptation of a framework that contextual data quality continuously changes and must be re-applied situationaliy, it is important that individual practitioners and data collectors be offered training on the application of a data quality system to ensure responsible use of data quality results. A needs and capability assessment highlighted lack of guidance on documentation and storage of DQC. Although datasets are documented, the DQC of the original data is either missing or partially stored in the background documentation. The lack of clear documentation of the DQC by generators is a hindrance to the interoperability of the data, since users must search through background documentation and or find original documentation of data in order to perform an evaluation on the contextual indicators. A method for data quality aggregation is proposed that extends earlier work (Rousseaux et al. 2001) to provide aggregate data quality scores for LCIA results. The use of data quality is recommended alongside, and not mixed with, quantitative uncertainty assessment. Reference [1] Weidema B, Bauer C, Hirschler R, Hertwich E, et al. (2013) Overview and methodology: Data quality guideline for the ecoinvent database version 3. The ecoinvent Centre, St. Gallen [2] Rousseaux P, Labouze E, Suh Y, Blanc I, Gaveglia V, Navarro A (2001) An overall assessment of life cycle inventory quality. INT J LIFE CYCLE ASS 6 (5):299-306

MO398 Long term effects of three different silver sulfide nanomaterials, silver nitrate and bulk silver sulfide on soil microorganisms and plants

Nanomaterials (NM) will enter the environment via diverse pathways. Sewage sludge for example is repeatedly applied as fertilizer on farmland due to its high nutrient content. This may lead to a significant increase in the occurrence of NM, particularly heater type and shower flow rate due to returning digestate from biogas plants to the field used for energy production. The results of the laboratory experiment over 180 days reflect the findings of the lysimeter study. For Ag-NM and CeO2-NM the results indicate that a hazard assessment based on data from laboratory tests is acceptable.

MO399 Site-specific N-emissions of rapeseed cultivation in Germany
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Globally growing population increases the demand for food, which should be produced as efficiently as possible but also environmentally friendly as possible. Simultaneously climate change requires the reduction of greenhouse gases (GHG) in order to keep the global temperature increase below 2°C. Germany has defined ambitious goals to reduce its GHG-emissions. The reduction targets are 40% by 2020, 55% by 2030, 70% by 2040 and 80-95% by 2050. Within the last 15 years, GHG emissions from agriculture have not decreased. Simultaneously the European Water Framework Directive requires a good status of water bodies, which is in particular regions in Germany not achieved. Winter oilseed rape (Brassica napus L., WOSR) is the major oil crop cultivated in Germany. Nitrogen field emissions are usually estimated using IPCC-emission factors that are not specific for the crop and associated with strong uncertainty. N2O field emissions are controlled by N fertilization and dominate the GHG balance of WOSR cropping due to the high global warming potential of N2O. The same applies for nitrate emissions that dominate the Eutrophication potential or ammonia emissions for the Acidification potential of WOSP when organic nitrogen fertiliser is applied. To address these issues and support decision makers, our project aims to educate specific emissions factors for WOSR and that can be included in the regional life cycle assessment studies for WOSR cultivation. Thus, field experiments were conducted to increase the data basis and subsequently derive WOSP-specific emission factors. Furthermore, the project strives to develop robust but also generalisable statements about nitrous oxide emissions and ammonia volatilisation due to returning digestate from biogas plants to the field used for state-of-the-art agricultural methods. The results will support the environmental assessment of WOSR production including additional aspects such as fertilizer use efficiency.

The environment as a reactor determining fate and toxicity of nanomaterials (P)
difference in the toxicity of the AgNMs. A realistic exposure scenario was chosen. The five test materials NM-300K, previously sulfidized NM-300K, a nanoparticulate Ag$_2$S, and bulk Ag$_2$S were added with an influent concentration of 1 mg/L and AgNO$_3$ with an influent concentration of 0.5 mg/L into the denitrification of a simulated STPs continuously for 10 days. The sewage sludge of each treatment was dewatered and the biosolids were mixed with soil. After 0, 60, 90, 120, 180 and 360 days, the release of Ag by the digestive gland was investigated (Avena sativa) and the substrate induced respiration (SIR, OECD 217) were observed. In addition, after 60 days of aging of the AgNM in the test soil a sub-sample was taken from each treatment and a chronic plant test was carried out (Avena sativa) and both the roots and the shoots were examined for an uptake of the Ag. We found an increasing inhibition of the ammonia oxidizing bacteria (AOB) from day 60 until day 180 for all tests. Bacterial activity in the digestive glands, but NPs were also detected when the soil was aged only for 60 days. This points to the formation of secondary NPs inside the organism. In vitro digestive juice model does not entirely represent the expected dissolution rate of NPs that was concluded from in vivo exposure. The usefulness of terrestrial isopods as models to assess the transformation of NPs will be discussed. This work was funded under NanoFase project (grant agreement No 646002).

**MO398**

**Energy reserves and respiration rate in the earthworm Eisenia andrei after exposure to zinc in nanoparticle or ionic forms**

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The energy budget is an indicator of the organisms’ overall condition and the changes in the energy reserves and/or energy consumption rate have been used as biomarkers of toxic stress. To understand better the effect of different forms and concentrations of Zn and possible costs connected with the effective Zn regulation by the earthworms, Eisenia fetida, between two conditions: Ko = 10 Kd (ZnO-NPs in Lufa 2.2 soil for 21 days (uptake phase), followed by 14 day elimination phase in clean soil), and Pb = 10 Kd (ZnO-NPs in Lufa 2.2 soil for 21 days (uptake phase), followed by 14 day elimination phase in clean soil). Two tests were conducted for both ZnCl$_2$ (250 and 500 μg Zn g$^{-1}$ dry soil) and ZnO-NPs (500 and 1000 μg Zn g$^{-1}$ dry soil), corresponding to EC$_{50}$ and EC$_{90}$ for reproduction, plus control without added Zn. The results suggest that the earthworms are able to regulate internal Zn concentrations efficiently, regardless of its form and concentration, and without any serious impact on their energy reserves. Sugar content was the only energy reserve component which was significantly lower in 1000 ZnO-NPs than control (p < 0.05) in the uptake phase. The total available energy reserves (Ea) and protein contents did not differ significantly between treatments but significant effect of day of exposure was found (p < 0.0003). Neither treatment nor the exposure day affected the lipid content in the uptake phase. In the elimination phase, no treatment or time of exposure had an effect on Ea and protein contents. The whole-organism respiration rate (measured as oxygen consumption) was not affected by Zn treatments in any of the two toxicokinetic phases. The results for the whole organism respiration rate will be additionally compared with those for the respiration rate measured at the cellular level as an electron transport activity, which is probably more prone to rapid temporal changes in conditions - as is the case for most biochemical biomarkers. The relationship between the two conditions: Ko = 10 Kd and Pb = 10 Kd metabolism (i.e. respiration rates measured at different levels) or energy budget (i.e. all energy reserve components) and internal Zn concentrations will be discussed. This study was supported by the National Science Centre, Poland (2015/17/N/NZ8/01576).

**MO399**

**Evaluating the Cellular & Humoral Immune Responses of the Terrestrial Isopod, Porcellio scaber, to Gold Nanoparticles**

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Gold nanoparticles are popular due to their unique physical and chemical properties. They can be synthesised and the myriads of potential uses they could have, which includes drug delivery systems, cancer therapy and biosensors. It is inevitable that these nanoparticles (NPs) will find their way into the environment and therefore the possible effects they could have need to be evaluated. In particular, it is anticipated that organisms may recognise NPs as “foreign” and respond by modulating their immune system. To date, only a few studies have dealt with this issue. The aim of this study was to investigate the question of whether or not the addition of gold NPs alters the immune response of the terrestrial isopod, Porcellio scaber. These organisms are well-studied and have previously been used as models for environmental toxicity. As the immune system is an early responder to foreign matter, studying it in conjunction with traditionally used parameters of toxicity can give more information into the possible effects these particles may have. This manuscript used two types of NPs: one without any coating and another with a PVP coating, both of which were approximately 26nm. For the ingestion route, animals were fed gold NPs for 14 days. During this time the feeding, defecation and survival rates of the animals was recorded. After 14 days, hemolymph was removed and the number, viability and proportion of hemocytes were counted. Along with the cellular tests, the humoral side of the immune system was studied by measuring the activity of the phenoloxidase pathway, which is associated with melanisation and wound healing, in the hemolymph. The levels of immune markers, glutathione S-transferase and soluble actylcholinesterase, were also assayed. As the gut is thought to impede the NPs’ ability to journey into the
MO402 Toxic Effects of Silver Nanoparticles and Its Transformation Product in Soil Applied with Biosolid
E. Topuz, I. Koyuncu, Istanbul Technical University / Environmental Engineering Biosolids, which are produced as a result of biological wastewater treatment, need to be managed as a separate waste stream. Land application of agricultural land, as a resource of nutrients and organic matter, is encouraged under the “Resource Efficiency Roadmap of Europe” [1]. However, the presence of contaminants in biosolids such as engineered nanoparticles can cause concerns. Total Ag concentrations in biosolids can be up to 195 mg Ag/kg dry soil in biosolids according to Johnson et al. [2] which is close to observed EC50 concentrations of AgNPs at AgNP (AgNPs) [3]. Moreover, AgNPs are mostly transformed to Ag sulphide nanoparticles (AgS-NPs) due to the reducing conditions present in the wastewater treatment plant (WWTP) [4]. Recent studies suggest the possibility of AgNP residuals because of the partial sulfidation of AgNPs [5]. Land spread of biosolids might lead to the transfer of AgNPs and AgS-NPs to the soil which could pose harm to soil organisms. Hence, this study aims to investigate the toxic effect of AgNPs, AgS-NPs, and their transformation product, Ag$_x$SNP, on soil ecosystem in the case of their spread via biosolid application. Polyvinylpyrrolidone coated AgNP (AgNP-PVP) and AgS-NP are tested in order to investigate their possible different effects on the survival and reproduction of Enchytraeus fetida. MO403 Short-term induced molecular stress responses in coelomocytes of Eisenia fetida earthworms in vivo exposed to silver nanoparticles s, curieuses, CONICET PRIET UNEU; N. García Velasco, E. Urionaharreentxea, University of the Basque Country UPV/EHU; M. Saenz, PRIET CONICET, National University of Luján; M. Soto, University of the Basque Country UPV/EHU; W. Di Marzio, CONICET-PRIET / PRIET
In recent years the production of nanoparticles (NPs) has increased massively. The subsequent release of NPs into the environment has raised questions regarding their potential ecological risk in soil, water and air. Silver nanoparticles (Ag-NPs) have the highest degree of commercialization due to their high thermal and electric conductivity, high catalytic activity, and powerful antimicrobial properties. Eisenia fetida is a model specie in soil toxicity studies and has been broadly used due to its sensitivity to different toxicants at different levels of biological organization. The main aim of the present investigation is to understand the effects produced by AgNPs (5,082 nm sized and PVP-PEI coated) in comparison with the soluble form of the metal (AgNO$_3$) at molecular level in coelomocytes of E. fetida at different exposure times. E. fetida were in vivo exposed to different concentrations of Ag-NPs and AgNO$_3$ (0.05 and 50 mg Ag/kg soil) through OECD artificial soil for 1, 3 and 14 d. Then, the transcription levels of selected genes associated to oxidative stress (Catalse) and metal detoxification (MTs-metallothionein) were determined in coelomocytes extruded from exposed earthworms. In addition, the enzymatic activity (Catalse) and protein content (MTs) were quantified. The responses varied significantly among days, exposure concentration and Ag form. Exposure to Ag-NPs led to...
significant induction of CAT at day 1, followed by an increase in its transcription levels after 3 and 14 d of exposure. Similarly, exposure to AgNO₃ induced the transcription of CAT at day 1 but at day 14 a downregulation was observed. The CAT activity increased at both treatment and exposure times (1 and 3 d). After 14 d of exposure, CAT activity was inhibited at the highest concentration tested. The highest increase of MTs at protein level was observed after 3 d of exposure. Our results indicate that short-term exposures to Ag NPs induced early molecular stress responses (MT induction and oxidative stress) in coelomocytes that preceded other responses at higher levels of biological organization. The responses in translational level in *E. fetida* tissues were according. The study indicates the importance of using integrative biomarkers for the evaluation of the potential risk of Ag-NPs in soils.

**MO404**

Effects of Cerium Nanoparticles with deferent surface-charge in coelomocytes of *Eisenia fetida*

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The majority of nanomaterials (NMs) used in commercial applications are likely to enter the wastewater stream and reach wastewater treatment plants. In many countries, wastewater effluent and sewage sludge are discharged in aquatic environments or applied on agricultural land, however, the transformation of the particles and their potential impact on human health is poorly understood. Recent studies have shown high association of NMs with sewage sludge, therefore soils can be a sink for NM pollution making terrestrial organisms vulnerable. The main aim of the study is to understand the transformation of NMs during wastewater treatment processes and to evaluate the potential environmental hazard of aged particles compared to pristine NPs. In this study, cerium nanoparticles (primary immune cells) isolated from the epigeic earthworm *Dendrobaena veneta* are used as a model to assess the effects of Ag and TiO₂ NPs. Initial investigations focus on Ag (PPV coated, 25 nm, nanoComposix) and TiO₂ particles (uncoated anatase, nominal primary size of 5 nm, NM-101, JRC) and their mixture, to better understand their uptake, interaction with coelomocytes and subsequent cellular effects. Moreover, a lab-scale wastewater treatment system is used to study the transformation of Ag and TiO₂ NPs through biological wastewater treatment processes, and the potential effects of the aged particles through biosolids application is evaluated. Extensive characterization of the particles in exposure media is performed with dynamic light scattering (DLS), single particle-Inductively Coupled Plasma Mass Spectrometry (sp-ICP-MS) and transmission electron microscopy (sp-TEM), while chemical markers of exposure (sp-ICP-MS) are measured on the sludge containing Ag and TiO₂ NPs. The effects of the pristine and aged particles on the metabolic activity, lysosomal integrity, reactive oxygen species formation, immune response and coelomocyte population are assessed. Moreover, nanoparticle uptake and intracellular localisation are evaluated with TEM and sp-ICP-MS.

**MO407**

Differential biomarker responses of *Daphnia magna* to pristine and wastewater borne silver nanoparticles

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The increasing use of silver nanoparticles (AgNPs) as antibacterial agents in a variety of products have raised the need to assess their environmental impact. The ever-growing application of AgNPs leads to their introduction into wastewater treatment plants (WWTWs) via sewer systems. During treatment, AgNPs are mainly retained in sewage sludge but part of transformed AgNPs is released into the environment. This study aims at investigating the effect of pristine and wastewater borne AgNPs on bioassays (C. elegans, *D. magna*) and *D. magna* responses to wastewater. The dispersants used in sludge (Surfynol® and Emiet®) are evaluated. Extensive characterization of the particles in exposure media is performed with dynamic light scattering (DLS), single particle-Inductively Coupled Plasma Mass Spectrometry (sp-ICP-MS) and transmission electronic microscopy (sp-TEM) to evaluate the potential environmental risk of AgNPs in wastewater treatment plants. The effects of the pristine and aged particles on the metabolic activity, lysosomal integrity, reactive oxygen species formation, immune response and coelomocyte population are assessed. Moreover, nanoparticle uptake and intracellular localisation are evaluated with TEM and sp-ICP-MS.

**MO405**

The uptake of pristine and aged silver nanoparticles by wheat, *Triticum aestivum*, in a soil exposure

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This study shows that (i) the response of biomarkers to used dispersing agent highlights the need for further study on its effects in organisms prior to its application, in order to understand the AgNPs behaviour in standardized test media, and (ii) there is a distinct biomarker response-pattern in daphnids exposed to WWTW effluent containing NM-300K and ASTM supplemented with pristine NM-300K. In
BioNano Interactions; I. Corsi, University of Siena / Physical, Earth and Environmental Sciences

Engineered nanoparticles (NPs), given the multitude of uses, can be released in aquatic environments, both intentionally and accidently. To date there is no shortage of studies concerning the environmental fate and ecotoxicity of widely used NPs, such as titanium and silver oxides. However, much less is known about NPs employed in novel cutting-edge applications as nanomedicines. Therefore, for aquatic environments we studied five biomedical NPs, namely aminated polystyrene (PSNH), europium doped-cerium oxide (CeO₂@Eu), carbon dot-doped silica (SiC@C), bare and polyethylene glycol-functionalized silica (SiO₂@B and SiO₂@PEG, respectively), and we assessed their behaviour and biological impacts in natural river (NRW) and seawater (NSW). Hydrodynamic sizes were monitored for 30 days by dynamic light scattering (DLS) and showed remarkable differences in NRW compared to NSW for both bare and PEGylated SiO₂ NPs. In fact, SiO₂ NPs dispersions were found to be stable in NRW, while an immediate instability was observed in NSW. PSNH@Eu and CeO₂@Eu and Si@C NPs did not show such a clear distinction between the two natural media, reaching micrometric sizes after 24 h. In order to address sedimentation phenomena, normalized derived count rates (DCR) were used to estimate the stability of suspended NPs in the two media. SiO₂@B and SiO₂@PEG NPs remained suspended in NRW until 10 days, while in NSW the sedimentation regime was weaker and hardly any signal was collected for suspensions after 24 h. On the contrary, no such difference was observed for PSNH@CeO₂@Eu and Si@C NPs, which completely settled within 24 h. NPs structural integrity was monitored as well over 30 days by means of spectrophotometric assays. SiO₂@B-based NPs bioaccumulation studies were examined in brown mussels (Mytilus edulis), which were conditioned before use with the test solution/dispersion to be filtered. PEG, respectively), and in silica (SiO₂@B and SiO₂@PEG NPs were carried out with the freshwater clam Corbicula fluminea. Silver nanoparticles (AgNPs) and TiO₂-NPs were chosen as model particles. The aim of this project was to detect and characterize MNMs in the tissue of animals collected at the end of the exposure period. For the analysis and characterization of nanoparticles in the tissue samples, two promising analytical methods were applied: (i) single particle inductively coupled plasma mass spectrometry (spICP-MS) as well as (ii) resolution transmission electron microscopy (TEM) imaging, while PSNH maintained an intact structure in NRW and NSW. Finally, algal growth inhibition tests were performed using freshwater and marine microalgae (OECD, 1994). PSNH and CeO₂@Eu were toxic in the OECD synthetic freshwater media only, while the remaining NP types did not show any sign of toxicity. A significant p < 0.05 reduction was observed in the PEGylated NPs in the tests performed in the two media, while no toxic effect was observed on the algae. Based on these results, the tests in NRW, while again no toxicity was confirmed in NSW. Altogether, our results provide a realistic insight in the fate and toxicity of diverse NPs, also highlighting the importance of testing complex natural matrices for a more realistic risk assessment.

MO409 Development of a method for the analysis of nanoparticles in the freshwater clam Corbicula fluminea

B. Meißenahr, Fraunhofer Institute for Molecular Biology and Applied Ecology IME / Ecological chemistry; B. Knopf, Fraunhofer Institute for Molecular Biology and Applied Ecology IME / Ecological chemistry; N. Schröder, Fraunhofer Institute for Molecular Biology and Applied Ecology IME / Ecological chemistry; S. Kühr, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology IME / Ecological chemistry; B. Schlechtriem, Fraunhofer IME / Department Bioaccumulation and Animal Metabolism Testing nanomaterials (NMs) under environmentally relevant conditions is an important aspect regarding the risk assessment of nanomaterials that enter the aquatic environment. The fact that sewage treatment plants (STPs) are the main pathway of NMs into the aquatic environment we developed a coupled test system using the effluents of model STPs in a chronic exposure test with the epibenthic amphipod Hyalella azteca, which is commonly used for ecotoxicity studies. Previous studies with this test system showed that silver (Ag) from silver NMs is accumulated by H. azteca exposed to model STP effluents. However, the pathway of Ag accumulation, via ingestion of particulate Ag and/or biocorrosion of dissolved silver, is still unknown. To further elucidate the uptake pathway of silver from model STP effluent, two groups of H. azteca with five animals each were placed in a single test vessel. The two groups were separated by a stainless-steel strainer. One group was fed contaminated sludge from model STPs, loaded on glass fibre filters. The second group, located in the stainless-steel strainer, was fed uncontaminated control STP sludge with no direct contact to the test organisms. Based on the results of these experiments, the study was carried out with four replicated test vials with two groups of amphipods each. Water samples were taken within the strainers to measure the silver content in the media and to prove that the animals fed control sludge were not in contact with Ag NMs potentially released from the contaminated sludge. After an exposure period of 7 (21) days Ag content of the water and animal samples collected at the end of the exposure period was measured by ICP-MS or ICP-OES to determine the accumulation of Ag in both groups. The presence of NMs in the animals was examined by high-resolution transmission electron microscopy (TEM) and methods of correlative microscopy. The derived accumulation factors and the results of the TEM investigations allow to evaluate the contribution of particulate and dissolved Ag to the accumulation of Ag from STP effluent.

MO412 Ecotoxicity of silica and silver nanoparticles (ENPs) on hyporheic copepods as a function of their bioavailability by dissolved organic matter (DOM) and water hardness of environmental samples

S. Marzo, CONICET; M. Saenz, PRIET CONICET, National University of Luján; D. Galassi, Universita dell’Aquila; J. Alberdi, National University of Luján; T. Di Lorenzo, Instituto de Ecosystem Study CNR The development and production of engineered nanoparticles has increased the potential for interactions of these nanomaterials with aquatic and terrestrial environments. ENPs are one of the commonly used particles in nanotechnology-based products. They are used in a wide spread application such as chemical industry, cosmetics, medicine and agriculture. The ENPs predicted concentrations using market study production estimates based on life cycle release models and measurements, in biosolids, was 5 - 123 and 0.02 – 2.01 mg/kg and, in
liquid effluents 0.03 - 6.74 and 0.003 – 0.26 g/L for Si and Ag NPs respectively. Environmental exposure modes have shown that soils and sediments can provide important reservoirs of these nanomaterials, especially in the presence of high concentrations of DOM. These such as the humic substances found in water, sediment, and soil, are ones of the substances capable of interacting with ENPs. To understand and assess the effects of NPs on the environment, should be well established quantitatively the concentration-response relationships. Also, to know how water treatment processes can regulate the fate of these nanomaterials and, thus their toxicity. Designed studies are therefore required in order to understand the fate, transport, stability, and toxicity of nanoparticles. By the other hand, there are not many studies about the effect of ENPs on hyporheic copepod species and less related with DOM concentrations. The hyporheic zone is a region underneath streambed that integrates surface and groundwater. Its location is central to biogeochemical linkages between the riparian zone, dissolved nutrients, and benthic biota. Even if in this DOM sources are relatively constant, biogeochemical processing within the hyporheic zone resulted a DOM pool that is temporarily dynamic regarding its composition and concentration. In this study we evaluated how DOM concentrations and water hardness are related with the acute ecotoxicity of Si and Ag NPs on the survival of Metacyclops gracilis, a widespread hyporheic species. Toxicity of AgNPs was related with DOM concentrations and showed a non-significant Beta for water hardness. On the contrary, for SiNPs, DOM and water hardness quantitative relationships were negatively correlated with ecotoxicity on this freshwater invertebrate.

MO413 Long-term exposure of ZnO nanoparticles to freshwater microalgae cultivated in batch and semi-continuous mode A.F. Aravantinou, F. Andreou, I. Manariotis, University of Patras / Civil Engineering Nanoparticles (NPs) have always existed in the physical environment. The rapid development of commercial applications involving the use of a large variety of synthetic nanoparticles has resulted in the introduction of higher amounts of nanoparticles in the environment. As the use of NPs increases, their effect to the coastal food chain and ecosystems is crucial. The aim of this work was to investigate the toxic effect of zinc oxide (ZnO) NPs on freshwater microalgae in batch and semi-continuous feeding mode for longer period than the time used in typical toxicity tests. Scenedesmus rubescens was selected as model microorganism since it is a common freshwater microalgae. S. rubescens exposed to ZnO NPs concentrations varying from 0.081 to 810 mg/L for 28 days in batch mode conditions, while in semi-continuous mode it was exposed to 0.081 mg/L of ZnO NPs. The cultures were grown in modified Blue-Green 11 medium (BG-11). The effect of ZnO NPs was assessed in synthetic wastewater mimicking river conditions including growth rate, nutrient removal and lipid production. The toxic effect of ZnO NPs was estimated by the half maximum inhibition concentration (IC50) and the growth inhibition rate (%) according to OECD 201 guideline. The experimental results in the batch mode conditions revealed that microalgae growth was significantly affected by the exposure time and the NPs concentrations. Specifically, the results showed that after a period of time the microalgae were adapted in the presence of ZnO NPs and were more resistant. In the semi-continuous mode the growth of S. rubescens was greater in the presence of ZnO NPs, and the lipid content was higher.

MO414 Effects of sunscreen-derived TiO2 nanoparticles on freshwater and marine organisms. M. vanni-cus Siliva, UNICAMP / Institute of Biology; S. cadore, University of Campinas; G. umbuzeiro, School of Technology, UNICAMP / LAEG The relatively recent development of engineered Ag nanoparticles has expanded silver uses considerably. Silver nanoparticles (AgNP) tend to aggregate in the aqueous phase and settle to sediment surfaces exposing deposit feeding organisms. Amphipods, like Parhyale hawaiensis, are deposit-feeding species ecologically relevant, becoming an excellent model for ecotoxicology studies. In addition to the traditional toxicity studies, internal doses determination, for example in the haemolymph of an amphipod, can be carried out by using AgNP. We hypothesized the typical AgNP could be absorbed by the gut leading to a higher amount of Ag in the haemolymph when compared to feeding containing AgCl. Silver nanoparticles < 100nm (Sigma Aldrich) or elemental Ag (from AgCl, Sigma Aldrich) were incorporated into the fish feed. In contrast, particle concentrations of approximately 200 mg kg-1 of P. hawaiensis organisms (6 months) were placed individually into a plastic container (100 mL of reconstituted saline water) and fed on alternate days with control, AgNP, or AgCl amended feed pellets. After 1 hour of feeding, each organism was washed and placed into a new plastic container with clean salt water to ensure that the exposure was only via food. The amphipods were then starved during 7, 14 and 28 days. After exposure, the haemolymph was collected using a thin glass capillary, weighted and analysed. Three pooled samples of 4 organisms (2 females and 2 males) were tested per exposure concentration. The silver determination in haemolymph was carried out by a Graphite Furnace Atomic Absorption Spectrometry (GFAAS). A higher amount of silver in the haemolymph was absorbed from AgNP feeding, reaching 8.4±0.7 mg kg-1 in comparison to 3.7±0.1 mg kg-1 for AgCl at the longest exposure time. The increase of silver concentration was related to the exposure duration. Therefore, it appeared that ingested silver is more bioavailable to P. hawaiensis as AgNP than when it is in its salt form. Data strongly suggest that nanoparticles were uptake by the gut and distributed in the UV radiation.

MO415 Silver nanoparticles affect the early development of Tisbe battaglial: pristine vs aged particles A. Georgantzopoulou, Norwegian Institute for Water Research NIV; K.J. farkas, SINTEF Ocean / Environmental Technology; K. landau, Institute for Environmental Sciences Group of Environmental Landau / Institute for Environmental Sciences Group of Environmental Research; M. oliviero, University Parthenope; A. Philippe, Universität Koblenz-Landau / Institute for Environmental Sciences Group of Environmental Chemistry; E. Stahl, Norwegian Institute for Water Research; P.a. Carvalho, SinTef Materials and Chemistry; A. Booth, SinTef Ocean / Environmental Technology; A. Macken, NIVA / marine pollution Silver and titanium nanoparticles are used in numerous consumer products and applications and they are likely to enter wastewater streams, reach wastewater treatment plants and aquatic systems through wastewater and effluent discharge. Nanomaterials undergo transformation in different matrices altering their fate, behaviour, bioavailability and toxic potential that could differ greatly from the pristine counterparts. There are challenges on the detection and quantification of nanomaterials at environmentally relevant concentrations in complex media and matrices such as whole organisms. Moreover, studies on uptake and effects of transformed particles on marine species is largely lacking. The aim of the present study is to better understand the transformation of Ag and TiO2 NPs in wastewater and assess the uptake, bioaccumulation and subsequent developmental effects of pristine and transformed particles on the marine organism Tisbe battaglial. In this study the harpacticoid copepod Tisbe battaglial was selected as a relevant marine species and the effects on the naupliar development was assessed over an exposure period of 6 days. The mortality and the developmental stage of the organisms was assessed daily. Initial investigations focused on the study of Ag (BGE coated in 25 nm nanoComposix) and TiO2 particles (nominal primary size of 5 nm, NM-101, JRC) and their mixture. Moreover, the particles were aged in synthetic wastewater (for 4 hours under stirring conditions) and the effects of “aged” particles on the uptake, bioaccumulation and naupliar development was assessed. Extensive characterization of the particles in synthetic wastewater, seawater and exposure media was performed with dynamic light scattering (DLS), ultraviolet–visible spectroscopy (UV-VIS), single particle-Inductively Coupled Plasma Mass Spectrometry (sp-ICP-MS) and transmission electron microscopy (TEM). Moreover, nanoparticle uptake, bioaccumulation and intracellular localisation is evaluated with TEM and sp-ICP-MS. Results showed that the both particles types were stable in seawater. Ag uptake was related with DOM concentrations and showed a negative correlation with the exposure duration. Therefore, it appeared that ingested silver is more bioavailable to P. hawaiensis as AgNP than when it is in its salt form. Data strongly suggest that nanoparticles were uptake by the gut and distributed in the UV radiation.

MO416 Silver concentration in the haemolymph of a tropical marine amphipod fed with silver nanoparticles and silver chloride M. Vannucci-Silva, UNICAMP / Institute of Biology; S. Cadore, University of Campinas; G. Umbuzeiro, School of Technology, UNICAMP / LAEG The silver determination in haemolymph was carried out by a Graphite Furnace Atomic Absorption Spectrometry (GFAAS). A higher amount of silver in the haemolymph was absorbed from AgNP feeding, reaching 8.4±0.7 mg kg-1 in comparison to 3.7±0.1 mg kg-1 for AgCl at the longest exposure time. The increase of silver concentration was related to the exposure duration. Therefore, it appeared that ingested silver is more bioavailable to P. hawaiensis as AgNP than when it is in its salt form. Data strongly suggest that nanoparticles were uptake by the gut and distributed in the UV radiation.

MO417 Silver concentration in the haemolymph of a tropical marine amphipod fed with silver nanoparticles and silver chloride M. Vannucci Silva, UNICAMP / Institute of Biology; S. Cadore, University of Campinas; G. Umbuzeiro, School of Technology, UNICAMP / LAEG The silver concentration in the haemolymph of a tropical marine amphipod fed with silver nanoparticles and silver chloride was measured by GFAAS. A higher amount of silver in the haemolymph was absorbed from AgNP feeding, reaching 8.4±0.7 mg kg-1 in comparison to 3.7±0.1 mg kg-1 for AgCl at the longest exposure time. The increase of silver concentration was related to the exposure duration. Therefore, it appeared that ingested silver is more bioavailable to P. hawaiensis as AgNP than when it is in its salt form. Data strongly suggest that nanoparticles were uptake by the gut and distributed in the UV radiation.
haemolymph causing this increase in Ag content. More studies are required to verify the Ag form present in the haemolymph and how it will induce damage in the exposed organisms.

**MO417**

**Toxic effects of multi-walled carbon nanotubes on bivalves: comparison between functionalized and unfunctionalized MWCNTs**

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The use of carbon nanomaterials (CNMs) has increased rapidly in the last years, namely due to their important properties such as electromagnetic, optical, catalytic, mechanical, thermal, and pharmacokinetics. Currently, carbon nanotubes (CNTs) are one of the most important and commercially used CNMs. CNTs are hollow graphene cylinders that are microns to millimeters in length and can be divided in single-walled (SWCNTs) with a diameter of 0.7 to 3 nm, and multi-walled (MWCNTs) with a diameter of 10 to 25 nm. CNTs are engineered with a wide variety of core structures and surface functionalizations that change their chemical and physical properties to enhance their suitability for different industrial applications. However, despite of the large array of available CNT configurations, their impacts on aquatic organisms, especially on invertebrate species, are still limitedly known. To our knowledge, no information is available on how surface chemistry alteration (functionalization) of CNTs may impact the toxicity of these CNMs. The basic reason for this exposure (28 days) to unfunctionalized MWCNTs (Ni-MWCNTs) in comparison with functionalized MWCNTs (f-MWCNTs), by introducing polar groups such as carbonyl groups (-COOH) in order to achieve better dispersability in water, were evaluated in the Manila clams Ruditapes philippinarum, one of the most dominant bivalve of the estuarine and coastal lagoon environments. Alterations induced in clams’ oxidative status, neurotoxicity and metabolic capacity were performed. The results obtained clearly showing that both Ni-MWCNTs and f-MWCNTs were able to generate oxidative stress in the exposed clams and were also responsible for changes in organisms’ metabolism (expressed in alteration of energy reserves) and neurotoxicity induction in R. philippinarum, however greater impacts were caused by f-MWCNTs resulted better dispersed than Ni-MWCNTs, NM100 resulted better dispersed than NM101 at 100 µg/ml and both titania showed similar agglomerate sizes at 10 µg/ml. We found lack of cytotoxicity for all NPs. Slight direct DNA damage at 10 and 100µg/ml and slight oxidative DNA damage at the lowest concentration were induced by NM200. NM203 induced dose-dependent direct DNA damage statistically significant at 100 µg/ml and oxidative DNA damage at low concentrations. NM100 induced dose-dependent direct DNA damage and oxidative DNA damage at 1 and 10 µg/ml. Direct DNA damage, statistically significant at 10 and 100 µg/ml, and induction of oxidative DNA damage at 100 µg/ml were found for NM101. Both silica NPs induced slight IL-8 release at 100 µg/ml, NM203 induced also IL-6 release at 10 and particularly at 100 µg/ml (262.2 fold of control). Both TiO2 NPs induced slight IL-8 release at 100 µg/ml but only NM101 induced significant IL-6 induction at 100 µg/ml. The findings show higher genotoxic/oxidative and inflammatory effects for NM203 in respect to NM200, probably due to its higher surface reactivity determining a strong interaction with the proteins in the medium and higher protein-mediated cell interaction. The findings also show DNA damage for both TiO2-NPs and oxidative DNA damage for NM101, correlated with the proinflammatory IL-6 cytokine induction, probably due to its smaller size, higher agglomeration tendency and capacity to induce ROS. This study is partially financed by FP7-NANOReg project, Grant n. 310584.

**MO419**

**Transformations of engineered nanomaterials during wastewater treatment: the role of engineered surface coatings and the impact on environmental fate**

M. Surrenti, Oregon State University / School of Chemical, Biological, and Environmental Engineering; J.A. Nason, Oregon State University / Chemical Biological and Environmental Engineering; R. Kaege, Eawag - Swiss Federal Institute of Aquatic Science and Technology

Previous research has highlighted the importance of particle-particle interactions in controlling the environmental fate of engineered nanomaterials (ENMs). Yet our ability to accurately predict the outcome of these interactions within environmental systems is still limited. One obstacle is the inherent complexity of these interactions which depend on several factors, including the properties of the ENM. However, the properties of the ENM are dynamic and can be altered via myriad transformation processes (e.g., over-coating via natural macromolecules, surface coating displacement, etc.). When considering the pathways by which ENMs may be released to the environment, wastewater treatment plants (WWTPs) not only act as gateways controlling the release of ENMs but they may also serve as reactors adjusting the properties of the ENMs. Therefore, to improve our understanding of ENM interactions within environmental systems we must first understand the extent to which ENM properties are altered within WWTPs. 'The objective of this research is to develop a protocol that simulates the transformations or ‘aging’ ENMs experience within a WWTP. The initial focus is on the effect of the dissolved components within the wastewater medium and whether ENMs with initially dissimilar properties will have similar properties after aging. To accomplish this, 12 different gold nanoparticles (AuNPs) with different engineered surface coatings were selected as model ENMs. A series of batch reactors, each containing a sample from a different stage in a WWTP, were used to assess the impact of each stage on the ENMs. Each wastewater sample was first filtered to remove suspended solids and then dosed with a single type of model ENMs. Each reactor was mixed and aliquots were collected over time. The aliquots were then analyzed by a variety of techniques to investigate the effect of the media on the properties of the ENMs, including size, surface charge, stability/aggregate structure, and hydrophobicity. Future research will investigate the impact of the suspended solids and the overall effect of the transformations on the aggregation behavior of the ENMs upon their introduction to different environmental mediums (e.g., surface water). In simulating the discharge of aged ENMs to the environment, wastewater treatment plants (WWTPs) not only act as gateways controlling the release of ENMs but they may also serve as reactors adjusting the properties of the ENMs.'
the sediment and lack of dissolution (< 1% of spiked cerium was extractable with water). Transdermal uptake of AgNP was attributed to dissolved silver in the pore waters and uptake of soluble silver, potentially through localised dissolution of particles at the worms’ surface.

**MO421**

**Examining the role of TiO2 nanoparticle surface transformations on transport and toxicity**

A.R. Deline, Oregon State University / Environmental Engineering; J.A. Nason, Oregon State University / Chemical Biological and Environmental Engineering

Titanium dioxide nanoparticles (TiO2 NPs) have great potential for use in a variety of commercial and environmental applications, including the photocatalytic treatment of contaminants. While processes like microbial inactivation and the generation of reactive oxygen species (ROS) have been studied under a variety of irradiation and water chemistry conditions, there exists limited mechanistic insight as to how these processes are related to the molecular-level surface transformations that may occur under different environmental conditions. This study is using surface-sensitive characterization techniques, including x-ray photoelectron spectroscopy (XPS), to examine the impact of UV irradiation, temperature, and water chemistry exposures on the metal oxide surfaces of both pure anatase and mixed anatase-rutile TiO2 NPs. Initial XPS studies of the impact of simulated solar irradiation revealed that solar irradiation resulted in a decrease in the aliphatic carbon present on the TiO2 surface and an increase in the oxygen-bonded carbon, with no observable effect on the oxidative properties of the metal oxide. Additional studies will be used to examine the ultraviolet and visible light effects for both TiO2 surfaces. To date, our Fresnel lens to increase the irradiation intensity and solution temperature. The relationships between these molecular-level surface properties and the extrinsic properties of the TiO2 NPs are being further explored using a suite of functional assays. Assays that have been optimized for the characterization of TiO2 NPs in this study include methylene blue dye degradation (photocatalytic activity), rose bengal dye photolysis (photodynamic), and thiochrome dye conversion (ROS generation). Ultimately, changes in the properties of the TiO2 NPs will be compared to larger scale environmental behavior, allowing for a better understanding of the specific role that surface structure plays in nanoparticle transport and toxicity.

**MO422**

**Influence of organic compounds on the sulfidation kinetics of copper oxide nanoparticles**

A. Gogos, Eawag, Swiss Federal Institute of Aquatic Science and Technology / Process Engineering, Particle Lab; A. Voegelin, R. Kaegi, Eawag - Swiss federal Institute of Aquatic Science and Technology

Organic compounds present in the environment can affect the transformation of engineered nanoparticles (ENM) in aquatic environments due to their influence on the kinetics and mechanisms of this transformation reaction. The focus of this study is to investigate the influence of organic compounds (DOM) on the kinetics and mechanisms of the sulfurization reaction of CuO nanoparticles (CuO NP). The sulfidation reaction and its kinetics have been described in detail. However, the influence of DOM on this reaction has not been investigated, yet. Due to their high content of biologically active substances, wastewater systems represent major sulfurizing compartments, which can influence the redox processes. In this study, three different organic model compounds (Bovine serum albumin (BSA), model protein), Alginate (model polysaccharide) and Polyacrylic acid (natural organic matter analogue)) and investigated their influence on the sulfidation of CuO NP. All experiments were conducted in solutions buffered to pH 8 at concentrations of 1.3 mM CuO and 4 mM HS−. Variable amounts of the organic compounds were added to reach final concentrations of 10, 100 and 1000 mg L−1. Reacted CuO NP were collected at selected time points and characterized using Cu K-edge X-ray Absorption Spectroscopy (XAS). In addition, selected samples were characterized using conventional electron microscopy (CSEM). M/Z values with high sensitivity (ICP-TOF-MS) were analyzed for both the size and number concentration in order to establish the transformation of CuO NP. MO423

**Evaluating the role of organic compounds on the sulfidation kinetics of copper oxide nanoparticles**

M.D. Montaño, University of Vienna / Environmental Geosciences; B. Gerstmann, A. Laycock, N. Tepe, T. Hofmann, F. von der Kammer, University of Vienna / Department of Environmental Geosciences

The advent of single particle ICP-MS (spICP-MS) has helped advance the field of nanotechnology, specifically at concentrations and in matrices that are environmentally relevant. However, the concentration of naturally occurring nanoparticles (NPNs) and nanomaterials far outweigh the expected releases of engineered nanoparticles (ENPs), making their detection by single element spICP-MS and their subsequent risk assessment a challenge. The introduction of ICP-time-of-flight-MS (ICP-TOF-MS) is potential to overcome these challenges, as elements are detected quasi-simultaneously at dwell times of 4µsec, covering nearly the entire atomic mass range (7-250 m/z). By examining differences in the chemical composition on a particle-by-particle basis, NPNs and ENPs can be differentiated, and geochemical processes occurring at the nano-scale can be explored on an individual particle basis. In order to establish this technique, several studies and analyses, involving different particle samples and sizes, were performed. Samples were analyzed using traditional spICP-MS (with quadrupole mass filtering) and with spICP-TOF-MS. The precision and accuracy for particle sizing and counting were evaluated for each technique for a range of elements to explore the capabilities and potential limitations of these techniques as to how these processes are related to the molecular-level surface transformations that may occur under different environmental conditions. This study is using surface-sensitive characterization techniques, including x-ray photoelectron spectroscopy (XPS), to examine the impact of UV irradiation, temperature, and water chemistry exposures on the metal oxide surfaces of both pure anatase and mixed anatase-rutile TiO2 NPs. Initial XPS studies of the impact of simulated solar irradiation revealed that solar irradiation resulted in a decrease in the aliphatic carbon present on the TiO2 surface and an increase in the oxygen-bonded carbon, with no observable effect on the oxidative properties of the metal oxide. Additional studies will be used to examine the ultraviolet and visible light effects for both TiO2 surfaces. To date, our Fresnel lens to increase the irradiation intensity and solution temperature. The relationships between these molecular-level surface properties and the extrinsic properties of the TiO2 NPs are being further explored using a suite of functional assays. Assays that have been optimized for the characterization of TiO2 NPs in this study include methylene blue dye degradation (photocatalytic activity), rose bengal dye photolysis (photodynamic), and thiochrome dye conversion (ROS generation). Ultimately, changes in the properties of the TiO2 NPs will be compared to larger scale environmental behavior, allowing for a better understanding of the specific role that surface structure plays in nanoparticle transport and toxicity.

**MO423**

**Evaluating spICP-TOF-MS for Exploring Environmental Nano-scale Processes**

M.D. Montaño, University of Vienna / Environmental Geosciences; B. Gerstmann, A. Laycock, N. Tepe, T. Hofmann, F. von der Kammer, University of Vienna / Department of Environmental Geosciences

**MO424**

**Assessing potential risks of Nanodrugs and their delivery systems in fish using Light Sheet Microscopy**

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Nanotechnology applications have increased dramatically in recent years including in the pharmaceutical sector. The unique properties of nanoparticles have been exploited in medicine in both drug development and drug delivery. Their small size and capability for manipulation and functionalisation allows for great improvements in drug efficacy. Nanomedicines can cross a wide range of biological membranes and barriers (including the blood brain barrier facilitating the diagnosis and treatment of life threatening diseases such as cancer. Although nanotechnology may help to reduce the toxicity and side effects of drugs, the actual carriers themselves may also have the potential for inducing toxic effects, depending on their composition. This raises the need for safety evaluations of these drug delivery systems both in patients, but also with respect to their potential for environmental impact. Very little is known regarding the potential impacts associated with the release of these nanodrugs or their carrier systems into the environment, although some studies have been begun to investigate the potential toxic effects of various nanoparticle shapes and coatings in aquatic organisms. Using various sizes of gold nanoparticles (between 10 and 100nm in diameter) with a non-reactive methyl polymer and fluorophore coating, we have traced their uptake and tissue partitioning using a casper mutant zebrasfish and light sheet microscopy. We have constructed a light sheet system based on the OpenSPIM platform, (SPIM - Selective Plane-Illumination Microscopy) which allows us to create 3D images and 4D videos in real-time. Using this rapid image acquisition technique we showed a size selective uptake of the nanoparticles into the kidney and minimal uptake in other organs. Deposition studies indicated a steady loss of the gold nanoparticles from the pronephric kidney over time. We also investigated for biological responses using specific zebrafish transgenic lines for oxidative stress and kidney function. We are now investigating the effect different coatings and functionalisations have on the uptake and distribution of gold nanoparticles in the larval zebrafish ultimately with the aim of beginning to define the potential for this important new group of medicines for having an environmental impact on fish.

**MO425**

**SETAC Nanotechnology Interest Group**

C. Svendsen, CEH, Wallingford / Pollution and Ecotoxicology

**Hydrophobic Chemicals and Mixtures: Reliable Investigations on their Environmental Fate and Effects (P)**
Effect of ageing on polymeric aromatic hydrocarbon composition of biochar

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The influence of ageing on biochar properties has been investigated by comparing three biochars that were artificially aged by either H2O thermal oxidation or horseradish peroxidase enzymatic oxidation. In addition, a field-aged counterpart for one of the biochars was recovered from an agricultural field site, four years after application. Biochar bulk properties showed only minor changes following both artificial and field ageing, indicating high biochar stability. Concentrations of the 16 US EPA PAHs were measured in all of the biochars and a contaminant trap was used to investigate the effect of ageing on their bioaccessibility. The concentrations of total and bioaccessible PAHs ranged from 4.4 to 22.6 mg/kg and 0.0 to 9.7 mg/kg, respectively. Concentrations of the 16 US EPA PAHs decreased following field ageing, but the proportion of low molecular weight PAHs increased. The observed changes in PAH composition with field ageing can partially be explained by uptake from the surrounding soil. In addition, size discriminatory intra-biochar transfer processes also contributed to the changes in PAH composition. To better understand changes in PAH composition with ageing, an additional broad range of alkylated PAHs was also analyzed in selected samples. Our results show that the tested artificial ageing protocols are unable to approximate the changes in PAH composition resulting from field ageing. Nevertheless, total and bioaccessible PAH concentrations decreased for both artificially and field-aged biochars, indicating that PAH release decreases with ageing. Therefore, well-produced biochars that meet European Biochar Certificate (EBC) and International Biochar Initiative (IBI) quality thresholds for total PAH concentrations are unlikely to present a risk with regard to PAH release following field application. These results have recently been published (doi: 10.1039/C7EM00116A).

Development of a Method for Measurement Freely Dissolved Concentrations of Alkylated PAHs Using Solid Phase Microextraction with PDMS Fibers

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Several current use pesticides (e.g. cyhalothrin, cyfluthrin, cypermethrin) and their metabolites are still applied in California. The bioaccessibility of these compounds, which are resistant to both chemical and physical degradation but have a high affinity for soil or sediment particles and organic matter, is crucial for understanding their potential for leaching and translocation. The U.S. EPA narcosis model (Hawthorne et al., 2006) requires the measurement of 18 parent and 16 groups of alkyl PAHs. Therefore, the present study investigated the bioaccessibility of these compounds using a new method. The concentrations of the 16 US EPA PAHs decreased following field ageing, indicating high biochar stability. Consequently, the method was used to investigate the effect of ageing on the bioaccessibility of PAHs. The concentrations of total and bioaccessible PAHs ranged from 4.4 to 22.6 mg/kg and 0.0 to 9.7 mg/kg, respectively. Concentrations of the 16 US EPA PAHs decreased following field ageing, but the proportion of low molecular weight PAHs increased. The observed changes in PAH composition with field ageing can partially be explained by uptake from the surrounding soil. In addition, size discriminatory intra-biochar transfer processes also contributed to the changes in PAH composition. To better understand changes in PAH composition with ageing, an additional broad range of alkylated PAHs was also analyzed in selected samples. Our results show that the tested artificial ageing protocols are unable to approximate the changes in PAH composition resulting from field ageing. Nevertheless, total and bioaccessible PAH concentrations decreased for both artificially and field-aged biochars, indicating that PAH release decreases with ageing. Therefore, well-produced biochars that meet European Biochar Certificate (EBC) and International Biochar Initiative (IBI) quality thresholds for total PAH concentrations are unlikely to present a risk with regard to PAH release following field application. These results have recently been published (doi: 10.1039/C7EM00116A).

Field testing of a new calibration approach for silicone passive samplers: Comparison of the concentration ratio method using samplers of different thicknesses with the PRC approach

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Silicone passive sampling is a common method for sampling bioavailable concentrations of waterborne hydrophobic pollutants in the environment. Often, silicone samplers have to be used in kinetic mode and sampler calibration is unavoidable. Most commonly, exchange kinetics are derived from the release rates of performance reference compounds (PRCs) spiked into the sampler prior to usage. Unfortunately, PRCs can be expensive and are not always available for all compounds. Due to these challenges, a complementary calibration approach using passive samplers of different thicknesses has been developed and investigated in lab-scale experiments. This study describes the testing of the approach in situ off the field. The sampling location was a storm water retention pond collecting storm water runoff from a motorway. The pond has two basins, one of which is equipped with a Floating Treatment Wetland (FTW) for cleaning the runoff. Two sets of duplicate samplers with 3 different thicknesses were installed in the inflow, after the FTW and in the reference basin without a FTW. One set of samplers was taken out after three weeks and the second after five weeks. These were extracted and analyzed for PAHs, with the concentration ratios for the different thicknesses used to calculate the field dissolved concentrations. All samplers had been additionally loaded with PRCs, with the decreases also used to calculate the field levels. These were compared to the results from the concentration ratio approach, underlining its suitability as a complementary calibration method and its application domain.
MO431 PAHs in water and surface sediments from Douro River estuary and Porto Atlantic coast (Iberian Peninsula, North Portugal) — Risks for biota and human health?
M. João Rocha, ICBAS; U.Porto, CIMAR / CIMAR LA; J.L. Dores-Sousa, VUB / Department of Chemical Engineering; C. Cruziero, CIMAR / CIMAR LA, Porto, CEF FCTUC U.Coimbra; E. Rocha, ICBAS U.Porto, CIMAR / CIMAR LA The study shows the presence of 16 priority pollutants (PAHs) in surface waters from the Douro River estuary and nearby Atlantic seacoast. These areas bath Porto and Gaia cities, widely known by the production/export of the famous Porto wine. This area, besides being highly industrialized, also holds an oil refinery, an important harbour, intense maritime traffic, and recreational marinas. For this study, water samples were taken from four strategic sampling sites, at six different times of the year. These samples were extracted by ultrasound technique (suspended fraction) and solid-phase extraction (dissolved fraction), before their quantitative analysis by gas chromatography—mass spectrometry (GC-MS). Data showed the presence of all analysed PAHs in all samples, which global amounts (Σ16PAHs) were extremely high in both analysed matrices and at all sampling sites. In fact, average concentrations attained ≤ 52 μg/g dry weight (dw) in surface sediments and ≈ 0.45 μg/L in water. The occurrence of PAHs might be due to the local industrial activities, such as the oil refinery, oil handling, and the transport of pollutants on water. The results of the evaluated concentrations, the surveyed areas were classified as highly polluted by these organics, suggesting that both mutagenic and carcinogenic responses can occur in both humans and aquatic animals living in these areas. This statement is supported by the measurement of carcinogenic PAHs for humans (group 1) dissolved in water (≤ 5%) and in surface sediments (≤ 6%) in biologically significant concentrations. These data are the first reported in this geographic area and can be used as a starting point for future control of the PAHs levels either locally either at the European scenario.

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Keywords: PAHs, carcinogenic, estuary, sea, monitoring

MO433 Occurrence and availability of PACs and total AhR agonists in contaminated soils - Combining in vitro reporter gene assay and chemical analysis with passive sampling and column leaching
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Polycyclic aromatic hydrocarbons (PAHs) are common contaminants at industrial sites, and occur as complex mixtures of thousands of PAHs and heterocyclic compounds (NSO-PACs) among others, collectively referred to as polycyclic aromatic compounds (PACs). The contaminant composition differ widely due to combination of many sources and the properties of the sites. Due to the high complexity in PAC-contaminated areas, current risk assessment of PACs is commonly based on chemical analysis of the 16 US EPA PAHs. Consequently, many PACs are unknown. There is an urgent need of improved and applicable analytical methods to assess environmental levels and fate of potential toxic PACs to evaluate risk to human health and the environment. An important concern regarding sites contaminated with PACs is the risk of groundwater contamination by release of the compounds from soils. The aim of this study was to investigate the occurrence of 77 PACs including PAHs, alkyl-PAHs, oxy-PAHs and NSO-PACs among total aryl hydrocarbon receptor (AhR)-agonists in soils from historical contaminated sites and to assess the availability of the compounds in the soils. A novel approach combining chemical (GC/MS) and biosynthetic measurements (H4IE-luc) combined with characterization of availability by use of a column leaching test and passive sampling was used. This approach allowed screening of potentially toxic metabolites of PACs in soils during remediation. The results show that chemical analysis of 16 US EPA PAHs to determine the degree of contamination of PACs in soils greatly overlooks toxicologically relevant PACs. Agonists in concentrations below the detection limit were not detected. The measured concentrations of PACs in all soils, indicating low availability of the compounds in soils. The leachable fraction was generally greater for more hydrophilic PACs, such as more polar PACs composed of two or three rings. Contribution of the analyzed PACs to the overall AhR-mediated activities detected in soils, leaches and passive samplers was pretty small and confirms presence of several other AhR agonists in soils. Only a small fraction of AhR agonists were available in soils, indicating an overestimation of the risk, if only total initial concentrations in soils would be considered in risk assessment. However, the results show that analysis of available fractions based on only 16 US EPA PAHs have the potential to underestimate the risk of the soils.

MO434 Verification of read-across for aquatic hazard properties of Petroleum Substances in REACH registrations
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Petroleum substances are examples of UVCBs (substances of Unknown or Variable Chemical Composition) and include complex mixture of Biological, Ecological, and/or Chemical materials (UVCBs). The complex chemical composition will varying depending on, amongst other things, the source of crude oil, the refinery processing and climate conditions. These substances present additional challenges when conducting environmental hazard and risk assessments under regulatory schemes such as REACH, and Concawe has developed bespoke models for these purposes. However, for purposes of hazard classification and labelling there is still a need for experimental aquatic toxicity test data on petroleum substances. Concawe substances have been organised into categories, based on similarities in refinery processes and physicochemical properties, resulting in a clustering of comparable chemical compositions and related hazard profiles. Applying read-across within a category is an established concept to fill in data gaps and to reduce unnecessary testing, and has been applied to available aquatic toxicity data to evaluate risk to human health and the environment. An important concern regarding sites contaminated with PACs is the risk of groundwater contamination by release of the compounds from soils. The aim of this study was thus to investigate (1) how to operate automated SPME on solid samples, (2) how to achieve equilibrium sampling for hydrophobic organics with a practical time span and (3) how to calibrate the new approach. Polychlorinated biphenyls (PCBs) served as model compounds and matrices included soil, sediment and sludge.

MO436 New approaches for determining solubility of volatile liquid chemicals
H. Birch, DTU Environment / Department of Environmental Engineering; L.N. Troelsen, Technical University of Denmark / Department of Environmental Engineering; P. Mayer, Technical University of Denmark / Department of Environmental Engineering
Water solubility is a fundamental parameter in environmental risk assessment of chemicals, and is theoretically a simple parameter to determine. For hydrophobic chemicals in the liquid state, the main challenge is to establish equilibrium between the pure liquid phase and the water phase within a reasonable time frame, while still allowing for any solute disappearance or droplet formation. A slow, drop-by-drop method for solubility measurements has previously been developed for this purpose, however it is time consuming as it requires weeks to equilibrate. In this work, two new approaches were used for solubility determinations. Both methods were originally developed for toxicity testing at the saturation level. Both approaches avoid direct contact between the pure substance and the water, thus minimizing the risk of droplet formation. The first approach uses passive dosing from a saturated silicone polymer in order to saturate the water, while the second approach equilibrates the water with the pure phase liquid through the headspace. Equilibrium time in the range of minutes to hours is expected for the two methods. Four liquid hydrophobic
chemicals within the logKow range of 4.4-8.6 will be included in the study, and results from both methods will be compared.

MO437 Headspace passive dosing for dose-response testing of volatile hydrophobic organic chemicals
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Constant and well-defined exposure is crucial for the toxicity testing of liquid organic chemicals with high Henry’s constants, which are prone to substantial evaporative losses. A simple and effective headspace passive dosing method was developed and then applied to control the exposure of the freshwater algae Raphidocelis subcapitata and the terrestrial springtail Folsomia candida to terpenes and alkane in toxicity experiments. The headspace passive dosing method applies a liquid partitioning donor placed in the headspace of the closed test vial for controlling exposure while avoiding direct contact and introduction of pure phase micro-droplets. Passive dosing from the pure liquid compound was applied for toxicity testing exactly at the solubility limit, and a dilution series of test chemicals prepared in purified vegetable oil served as donor for dose-response testing. The terpenes S-(-)-Limonene and a(+)-Pinene were tested in both the algal growth inhibition test and the springtail test. In addition, n-nonane, n-undecane and n-dodecane were tested on the algae, while iso-octane, iso-dodecane and n-dodecane were tested on the springtails. Our first results demonstrated that (1) the headspace passive dosing method is a simple yet effective way to control exposure to volatile hydrophobic organic chemicals and (2) that the method is straightforward to apply in algal growth inhibition and springtail toxicity tests. Further analyses of exposure parameters are in progress to better understand and quantify the resulting toxicity.

MO438 Application of biomimetic solid phase microextraction to characterize aquatic hazard of petroleum substances
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Environmental hazards of petroleum substances differ in response to variable substance composition. In response CONCAWE has initiated a comprehensive analytical program to extend analytical characterization of petroleum substances to further support hazard classification within and across petroleum substance categories. As part of this work, SPME and toxicity data for newly characterized substances (n=139), across approximately 10 major categories, were compared to historical data. New compositional data were used as input to PETROTOX to predict toxicity. In this study, the freely dissolved concentrations (C_{fw}) of the chemicals were determined using equilibrium with thin coatings of silicone on the inner walls of glass jars with subsequent solvent extraction. Total sediment concentrations (C_{total}) were determined using accelerated solvent extraction. While there is a wide range of pollutants that have been detected in sediments worldwide, traditional chemical analysis cannot cover all compounds and their transformation products. Therefore, in this study, the extracts were dosed into seven cell-based bioassays covering cytotoxicity, activation of metabolic enzymes (binding to the arylhydrocarbon receptor, AhR), specific, receptor-mediated effects such as estrogenicity (ERα); and adaptive stress response (oxidative stress, AR Ec32). Cytotoxicity was assessed in all bioassays and occurred occasionally. Moreover, four of the seven bioassays were selected as the “first line” assay in 2012, the preferred receptor for the arylhydrocarbon receptor gamma (PPARγ) and ERα. The activation of the AhR was by far most responsive and showed a distinct pattern across the sampling locations. The other three assays showed responses only at higher enrichment factors of the extracts, also revealing specific contamination patterns. A comparison between C_{fw} vs. C_{total} will enable assessing the actual risk (C_{real}) vs. the potential hazard of those chemicals that might be released in future scenarios (C_{bio}). The presented work calls for more detailed studies at specific sites and testing of additional endpoints with the aim of obtaining a complete picture of mixture effects caused by the freely dissolved and total concentrations of hydrophobic organic chemicals in sediments.

MO441 Bioaccumulation of hydrophobic organic compounds in aquatic biota: addressing current challenges for in tissue passive equilibrium sampling
E. Rojo-Nieto, Helmholtz centre for environmental research - UFZ / Department of Cell Toxicology; J. Koschorreck, Umweltbundesamt; m. muz, UFZ - Helmholtz Centre for Environmental Research / Cell Toxicology; A. Jahne, Helmholtz Centre for Environmental Research - UFZ GmbH / Cell Toxicology

Bioaccumulation of hydrophobic organic compounds in aquatic biota: addressing current challenges for in tissue passive equilibrium sampling
E. Rojo-Nieto, Helmholtz centre for environmental research - UFZ / Department of Cell Toxicology; J. Koschorreck, Umweltbundesamt; m. muz, UFZ - Helmholtz Centre for Environmental Research / Cell Toxicology

The occurrence of a range of historical and emerging hydrophobic organic contaminants in mangrove ecosystems in Singapore. In particular, the levels of synthetic musk fragrance compounds, polychlorinated biphenyls, organochlorine pesticides and polycyclic aromatic hydrocarbons were measured in mangrove sediments, clams and caged mussels. In addition, the freely dissolved concentration of these organic chemicals in water was assessed with silicone rubber passive samplers. Results showed that polycyclic musks are present in mangrove ecosystems, and can accumulate in the tissues of molluscs. In the present study, bioaccumulation factors (BAFW, wet weights) were calculated for all the samples/sites and log BAF_{fw} averaged 4.0±0.3, 4.4±0.3, 4.7±0.3, 3.9±0.7 and 4.3±0.4 for galactolide, trasoleide, phenanthrene, carboxylated and tolanide respectively. Overall, the empirical models fit reasonably well the bioaccumulation of polycyclic musks in both caged and native molluscs in tropical mangroves. The study of the bioavailability of hydrophobic compounds in highly dynamic environments such as mangroves can be sometimes intricate, and the usefulness of passive samplers and sentinel species such as bivalves was confirmed in the present study.

MO440 Effect-based characterization of mixtures of environmental pollutants in sediments collected between the Arctic and Australia
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There is a multitude of pollutants that combine persistent and hydrophobic properties. In aquatic environments, they are largely deposited in sediments. The amount and characteristics of the organic carbon determine how strongly they are present in sediments. The present study reveals that the freely dissolved concentrations (C_{fw}) of the chemicals were determined using equilibrium with thin coatings of silicone on the inner walls of glass jars with subsequent solvent extraction. Total sediment concentrations (C_{total}) were determined using accelerated solvent extraction. While there is a wide range of pollutants that have been detected in sediments worldwide, traditional chemical analysis cannot cover all compounds and their transformation products. Therefore, in this study, the extracts were dosed into seven cell-based bioassays covering cytotoxicity, activation of metabolic enzymes (binding to the arylhydrocarbon receptor, AhR), specific, receptor-mediated effects such as estrogenicity (ERα); and adaptive stress response (oxidative stress, AR Ec32). Cytotoxicity was assessed in all bioassays and occurred occasionally. Moreover, four of the seven bioassays were selected as the “first line” assay in 2012, the preferred receptor for the arylhydrocarbon receptor gamma (PPARγ) and ERα. The activation of the AhR was by far most responsive and showed a distinct pattern across the sampling locations. The other three assays showed responses only at higher enrichment factors of the extracts, also revealing specific contamination patterns. A comparison between C_{fw} vs. C_{total} will enable assessing the actual risk (C_{real}) vs. the potential hazard of those chemicals that might be released in future scenarios (C_{bio}). The presented work calls for more detailed studies at specific sites and testing of additional endpoints with the aim of obtaining a complete picture of mixture effects caused by the freely dissolved and total concentrations of hydrophobic organic chemicals in sediments.
lipid), have been selected to optimise silicone-based PSDs for sampling in lean tissues. For this study, silicone is used as common reference phase, with sampler relocations across the homogenated samples along the sampling period, as has been proposed by Rusina et al. [1], in order to avoid the local depletion of the sample in direct contact with the silicone, and both, kinetic and equilibrium approaches have been considered. The 7 indicator PCBs (28, 52, 101, 118, 138, 153, 180) have been selected as target compounds, covering a logKow range from 5.66 to 7.15. Reference: [1] Rusina TP, Carlsson P, Vrana B, Smedes F. 2017. Equilibrium passive sampling of POP in lipid-rich and lean fish tissue: Quality control using performance reference compounds. Environ. Sci. Technol., DOI: 10.1021/acs.est.7b03113.

MO442 Widespread occurrence of 4-Nonylphenol, BHT, and 2,4-DTBP in blue crab, Callinectes sapidus, megalopa in the northern Gulf of Mexico S. Chiasson, Loyola University / EEB; E.K. Grey, Governors State University / Division of science, mathematics, and technology; D.A. Grimm, Tulane University / Coordinated Instrumentation Facility; C.M. Taylor, Tulane University / Ecology & Evolutionary Biology. The blue crab, Callinectes sapidus, is an ecologically and economically important invertebrate species in the northern Gulf of Mexico (NGOM). The NGOM receives nearly 60% of drainage from the river systems in the continental United States. Blue crab megalopa collected over three years from multiple estuaries in the NGOM from Texas to Florida were tested for alkylphenol contamination using GC/MS. We found all the target compounds were present in megalopa but at levels that may not be an indication of a potential marine pollution but exhibits low toxicity. All these compounds have high partition coefficients, which explains their presence in animal tissue. Concentrations of NP, BHT, and DTBP are highly correlated in megalopa over time at most study sites, implying a common source. The relatively high NP concentrations detected in juvenile blue crabs across the NGOM suggests that management strategies of alkylphenols in the environment should be re-evaluated to mitigate potential sub-lethal effects of exposure to blue crab populations.

MO443 Real-time visualization and quantification of perylene bioaccumulation at single cell level x. guo, School of Environment, Beijing Normal University; X. Jin, Beijing Normal University; F. Bai, Peking University. Hydrophobic organic chemicals (HOCs) are of special ecological concern because they can be directly incorporated and bio-concentrated in living organisms. However, the effects of self-clustering of HOCs on their environmental behavior and toxicity are not well understood. In this study, single cell suspensions of E. coli were exposed to perylene, a common contaminant in marine pollution, to study the cell-to-cell heterogeneity. Perylene bioaccumulation in single bacterial cells and examine the cell-to-cell heterogeneity. Although with identical genomes, individual E. coli cells exhibited a high degree of heterogeneity in perylene accumulation dynamics, as shown by the high coefficient of variation (C.V. = 40). This remarkable heterogeneity was exhibited only in live E. coli cells. However, the bioaccumulation of perylene in live and dead S. aureus cells showed similar patterns with a low degree of heterogeneity (C.V. = 0.36). We found that the efflux systems associated with Tol C played an essential role in perylene bioaccumulation in E. coli, which caused a significantly lower accumulation and a high cell-to-cell heterogeneity. In comparison with E. coli, the Gram-positive bacteria S. aureus lacked an efficient efflux system against perylene. Therefore, perylene bioaccumulation in S. aureus was simply a passive diffusion process across the cell membrane. With the use of SMFM, the motion and distribution of perylene nano-clusters (PCNs) formed in water at very low concentration were visualized with high temporal and spatial resolution. Moreover, the transport of PCNs across the cell membrane was also real-time captured, demonstrating that they entered macrophage cells by endocytosis. Supplementing the well-recognized routine of passive diffusion through membrane lipid bilayer, the uptake of HOCs in the form of nano-clusters by endocytosis was proposed to be an additional but important mechanism for their uptake into living cells. HOCs distributing in the environmental systems in the form of nano-clusters, as exemplified by PCNs in this study, may have significant implications for understanding their environmental fate and potential toxicological effects.

MO444 Impregnation factors of freshwater fish by organic micropollutants in the Marne Hydrographic network N.F. Molbert, UPMC UMR METIS 7619 / Biogeochemistry; M. Cheveuille, EPHE / UMR METIS 7619; F. Alliot, EPHE / UMR Metis; R. Santos, HEPIA; J. Mouchel, UPMC UMR METIS CNRSUPMC, A. Goute, EPHE / UMR METIS Ecotoxicology faces the challenge of monitoring the levels of an increasing number of chemicals on biota. While persistent pollutants have been largely studied, several pollutants are metabolized, especially by vertebrates. Despite the higher toxicity potential of metabolites compared to their parent compounds, little attention has been given to metabolites. Several persistent micropollutant families (Polychlorinated Biphenyls (PCB), Organochlorine Pesticides (OCP)) and metabolizable ones (Polycyclic Aromatic Hydrocarbons (PAH), phthalates, pyrethroid pesticides), as well as their metabolites were measured in a free-living freshwater fish, the European chub (Squalius cephalus, N = 113) caught by electrofishing operations in the Seine watersheds. The occurrence of pollutants was characterized in muscles and their metabolites in bile and liver using gas chromatography (GC-MS/MS) and high performance liquid chromatography (HPLC-MS/MS) coupled to a mass spectrometer. Body burdens of pollutants in chubs were then explained according to the environmental (water surface and sediments) contamination and individual parameters (age, body length, health status and parasitic load). Despite restrictive legislation, persistent pollutants (OCP, PCB) were found in all environmental matrices and fish tissues. Phthalates were the most abundant chemicals, with concentrations in fish muscles in the range 41.6-2200 ng g⁻¹. Positive correlations were detected between the environmental pollution and the levels of persistent pollutants (PCB, OCP) in chubs, but not for the metabolizable chemicals, likely due to their rapid degradation and excretion. No correlation was found between micropollutant levels and health status of chubs, suggesting low ecotoxicological effects of these contaminants exposed in the Marne hydrographic network. Surprisingly, chubs infected by the acanthocephalan Pomphorhynchus laevis were less contaminated. In the study of 60 chub samples, NP, BHT and phthalates are widely used in plastic products, paints and coatings to improve the product stability against UV light. Furthermore, some UV stabilizers are approved as ingredients in personal care products like sunscreens to protect human skin against harmful effects of UV radiation. The pathways into the marine environment are either indirect by wastewater treatment plant discharges or direct by recreational activities like bathing and angling. Four benzene derivatives are classified as SVHC (Substances of Very High Concern) under the EU legislation REACH. Numerous others are currently listed under the European community rolling action plan (CoRAP) to be re-evaluated in the next years. Due to their chemical properties, most UV stabilizers accumulate in sediment (logKow > 3) and have potential for persistence or pseudo-persistence. Environmental data form coastal and marine lakes were insufficient and sparse. In the study of 60 sediment samples of the North and Baltic Seas were analysed for 19 commonly used organic UV stabilizers. The sample pretreatment and analysis was carried out as following: First, the samples were homogenized with sodium sulphate. Afterwards, extraction and clean-up was performed using an accelerated solvent extraction (ASE-350, DIONEX, Germany) method. For this, 22 mL stainless steel ASE cups were filled with the activated silica and approximately 5 g sediment that was spiked with appropriately isotopically labelled standards. The standards were extracted using dichloromethane for three 10 min-cycles at 100 °C. The extracts were solvent-changed to methanol and reduced in volume to 150 µL. The instrumental analysis was performed on a LC-MS/MS system (1290 Infinity coupled to 6490 triple quadrupole LC/MS, Agilent Technologies, Germany) equipped with an APPI-source and a C18 column (Eclipse Plus RRHD 1.8 µm, 2.1 x 150 mm, Agilent Technologies, Germany). Total Organic Carbon (TOC) analysis was carried out with aliquots of the freeze-dried samples using a LECO RC612 multichapse (Germany). This study shows levels of contamination and distribution of organic UV stabilizers in surface sediments of the North and Baltic Seas for the first time. Several substances have been identified in concentrations in the low ng/g dw range.

MO446 Is Lake Como a "uniform lake"? Information from its inhabitants (zooplankton and fish) M. Bonomi, University of Insubria (Como) / DiSTA; A. Buflo, F. Cappelli, Water Research Institute - Italian National Research Council IRSA-CN; R. Penna, University of Insubria; S. Polesello, Water Research Institute- CNR / Water Research Institute; S. Valsecchi, Water Research Institute - Italian National Research Council IRSA-CN; R. Bettinetti, University of Insubria / DiSTA Lake Como, a subalpine lake (Northern Italy), is an oligomictic lake, with complete water mixing occurring after particularly windy and cold winters. It presents a typical shape of an upside "Y" where a western, eastern and northern basin can be identified. In more detail, the western branch is distinctly separated from the rest of the lake by an underwater ridge, where the highest depth is measured (425 m at 237
Argegno), and does not present an outlet, resulting in a longer real water renewal time. On the other side, waters of the eastern branch are encouraged to flow towards south directly through the Adda River, which is also the main inlet in the northern branch. Western and eastern branches also present different level of trophic status. In the present work, we investigate if these main morphological features can lead to differences a) in zooplankton density and biomass, b) in the interactions between zooplankton and phytoplankton, and c) in levels of pollution between the pelagic areas of the two branches, evaluated along a seasonal sequence. Preliminary data tell that the taxa composition of the pelagic planktonic communities is the same in the basins but differences in density and biomass are highlighted. These differences are found in levels of contamination of legacy compounds (DDT, PCB) while there are not differences between the two branches in concentrations of perfluoralkyl substances (PFAS).

MO448
Kinetic Sorption and Bioaccumulation of Hydrophobic Organic Chemicals in Marine Plankton Food Chain
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Bioaccumulation and distribution of polycyclic aromatic hydrocarbons (PAHs) over different amounts of exposure time were investigated in the plankton food chain including phytoplankton and zooplankton. The simulated plankton food chain was used to study phytoplankton (Tetraselmis chuii), rotifers (Brachionus sp.), and copepods (Acanthocera sp.) in a controlled and a gas nutrition system with a steady supply of PAHs for 7 days in this study. The results show that PAH accumulation in plankton can be roughly divided into three sections: 0.2-1 hours, 1-24 hours, and 24-168 hours. The PAH concentrations in plankton varied greatly over the 0.2-1 and 1-24 hour time intervals, then approached study-state at 24-168 hours exposure. The low molecular weight PAHs (ACN, AC) were found at significantly higher levels compared to than in high molecular weight PAHs (PA and CY). The PAHs were found at significantly higher levels in phytoplankton, indicating that plankton might have selectivity towards PAHs. In principal component analysis (PCA), the plankton could be separated significantly into phytoplankton and zooplankton. Parts of the PAH accumulation found in rotifers and copepods were similar, demonstrating that PAH composition in plankton might be affected by trophic levels. All PAHs demonstrated significantly linear relationships between bioconcentration factor (BCF) and PAH hydrophobicity (log Kow) in plankton, however the linear difference regression slopes of log BCF and log Kow between phytoplankton, rotifer and copepod, suggested that the plankton have different pathways of PAH accumulation.

MO449
Do weathered multiwalled carbon nanotubes influence the distribution of the biocide triclocarban in a sediment-water system?
L. Benner, U. Piltowski, M. Hennig, H. Hohler, RWTH Aachen University / Institute for Environmental Research; A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics.

Multiwalled carbon nanotubes (MWCNT) are widely used nanomaterials in a variety of different products and processes, well-known for their high sorption capacity. Due to increasing usage and production, exposure to the aquatic environment either accidentally or via disposal of CNT-containing products might increase likewise. Weathering processes like radiation can alter nanoparticle properties and lead to changes in their environmental behaviour. The present study investigated the potential sink for carbon-based nanomaterials. Nevertheless, during their stopover in the water phase they may interact with water dissolved xenobiotics, and thus alter their environmental behaviour. The duration of environmental exposure leads to interferences during chemical analysis, hindering the assessment of toxicological effects. Thus, the analysis of PAHs in the present study are based on PAH standards. Additionally, the data show that PAHs have a significant impact on the aquatic environment.

MO450
When technical limits triggers risk assessment for non-biodegradable insoluble pharmaceutical molecule
B. Journel, E. Beltran, CEHTRA SAS; P. Adrian, CEHTRA.

Face of substances in the environment are driven by numerous factors. Among them, substance’s properties such as Henry’s constant (i.e. water solubility and volatility) and hydrophobicity (in terms of Kow and adsorption) have a non-negligible impact on how substance’s behaviour is modelled in the different compartments (soil, water, sediment, air), to predict exposure levels in risk assessments. Pharmaceuticals represent a specific category of substances as they are difficult to analyse, which requires special experimental results more subject to imprecision. Due to analytical difficulties, parameters such as water solubility and Kow, are often expressed as “lower than” or “higher than” and have no defined value. Additionally, model softwares such as EUSES impose maximum value for Kow and minimum value for water solubility as input parameters, whereas EU TGD spreadsheet allows to consider the experimental results.

MO451
Effect of environmental characteristics on the bioavailability of hydrophobic organic compounds to fresh water organisms from natural aquatic systems
L. Teunen, University of Antwerp; C. Belpaire, Research Institute for Nature and Forest INBO; R. Blust, L. Bervoets, University of Antwerp / Department of Biology (SPHERE Research Group).

Many aquatic ecosystems are under persistent stress due to influxes of anthropogenic chemical pollutants. High concentrations can harm entire ecosystems and be potentially toxic to humans. The European Water Framework Directive (WFD) obliges member states to monitor chemical compounds in surface waters and to set quality standards that are protective for the ecological integrity. Generally, most of the target chemical compounds are investigated in environmental samples. However, in the case of highly hydrophobic compounds, their very low water solubility precludes direct measurement in water, and thus alternative monitoring strategies are needed. Accordingly, the WFD has formulated biota quality standards (BQS) which refer to concentrations of compounds that have to be monitored in fish and invertebrates. In the present study we are investigating the reliability and relevance of BQS by studying the relationships between concentrations of hydrophobic compounds in environmental compartments (mainly in sediment) and concentrations in biota. Our study encompasses 22 field locations at which we are monitoring the concentrations of a set of hydrophobic organic compounds and total Hg in both sediment and biota (fish and mussels). In addition, some sediment characteristics, i.e. organic carbon content (TOC) and clay content are measured and water characteristics are monitored, i.e. pH, oxygen level and conductivity. For each of the measured compounds multiple regressions are being constructed to establish the links between the concentration of compounds in biota and in sediment. The interpretation will take into account dissolved concentrations (where feasible) as well as general water and sediment characteristics. Identification of robust links between the extent of bioaccumulation and sediment and/or water concentrations would strengthen the basis for use of surrogate monitoring methods.

MO452
Personal care products (PCPs) in the southeastern coast of Brazil: determination of the method and environmental occurrence
T. Combi, Instituto Oceanográfico da Universidade de São Paulo / Instituto Oceanográfico; R.C. Montone, Universidade de Sao Paulo / Oceanographic Institute.

The use of large amounts and a broad variety of chemicals that are potentially harmful to the environment (including persistent organic pollutants (POPs), pharmaceuticals and personal care products (PCPs)) is widely spread in the environment and sediment and biota. The existence of such a high number of compounds in the environment leads to interferences during chemical analysis, hindering the assessment of the occurrence and distribution of contaminants in environmental matrices. Thus, the development and optimization of analytical methods that can detect multiple classes of compounds at very low levels and the contamination assessment in marine and coastal areas are among the most complex and current issues in environmental chemistry. Possible adverse effects related to some groups of PCPs, such as potential to accumulate through the trophic food chain and in human adipose tissues, endocrine disruption and hazard to coral reef conservation.
have been recently detected. Consequently, their study has become a priority among the main bodies responsible for protecting public health and the environment, such as the European Commission and USEPA. However, the current knowledge about the occurrence and fate of PCPs is still scarce, especially in less developed or developing countries as Brazil. Thus, this study aims to evaluate the occurrence of PCPs in surface sediments of selected areas along the southern and southeastern Brazilian coast through the optimization and implementation of a state of the art methodology. Preliminary results obtained for surface sediment samples from São Paulo coastal areas through microwave-assisted extraction (MAE) and triple-quadrupole mass spectrometer analyses (GC-MS/MS) revealed the presence of UV-filters (especially octocrylene and EHMCS) and fragrances (tonalide and galaxolide). The next steps of this work include testing additional extraction methodologies, extraction solvents and clean-up procedures to improve the detection and quantification of these compounds. The final results of this work will provide the first extensive dataset on the occurrence, levels and fate of PCPs in the Southern Atlantic which will not only contribute with new and rather scarce data but also with valuable information for regional and global inventories.

MO453 IFRA Environmental Standards and RIFM Safety Assessment Program Advances Update for 2018
A. Łapczynski, RIFM / Environmental Science; D.T. Salvito, Research Institute for Fragrance Materials (RIFM) / Dept of Environmental Science; C. Gonzalez, IFRA
To assure safety of fragrance ingredients in consumer products, International Fragrance Association (IFRA) developed the fragrance industry’s self-regulatory safety program with the development of IFRA Environmental Standards for both risk and hazard in 2008. Fragrance material risk assessments for these Standards are incorporated in the Research Institute for Fragrance Materials’ (RIFM) testing program in coordination with its Expert Panel. To identify materials for risk assessment refinement, fragrance materials were screened using the RIFM Environmental framework in 2008 IFRA volume of use survey as reported for both Europe and North America. The Framework for this evaluation was published in Environment Toxicology and Chemistry (Salvito et al., 2002, 1301-1308). In addition, hazard assessment on these materials was also performed and reviewed. As a result nearly 3,000 materials were screened with preliminary risk quotients estimated to rank materials for risk assessment refinement in an effort to provide greater transparency to the IFRA Environmental Standards, RIFM reports the most recent results of these additional tests (for both risk and hazard assessments) at both the annual SETAC NA and Europe meetings. These studies include persistence testing (ready biodegradation tests and die-away studies), bioaccumulation, and acute and chronic aquatic toxicity. Incorporating these new data in a second tier risk and hazard assessment for these materials will also be presented.

MO454 Comparison of different sampling techniques for the identification fire effluents from low-density polyethylene burning
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Huge amounts of various polymers are being used in many fields with numerous benefits. However, their great ability to ignition and rapid flame spreading make these materials dangerous for human life and properties due to the release of highly toxic combustion products. The present work aims to investigate several methods of sampling and analysis of polyyclic aromatic hydrocarbons (PAHs) produced by controlled burning of low-density polyethylene (LDPE) using a toxicity tube furnace. Five different sampling methods were used: solid phase micro extraction (SPME), syringe, tedlar bags, sorption tubes, and gas-solution absorbers. The produced hydrocarbons were analysed by gas chromatography coupled to mass spectrometry with and without pyrolysis. The analysis of PAHs released from polyethylene combustion showed that emissions with a potentially negative impact on the human health and the environment are produced in significant concentrations. Among the tested techniques, the most convenient sampling method was that using syringe with a glass vessel which allowed detection of the highest amount of PAHs at both 800 and 600°C, then followed by SPME. On the other hand, the use of gas-solution absorber (midget impinger) showed poorer results. Regarding the use of tedlar bags and sorption tubes, they did not give satisfactory results. Several carcinogenic or possibly carcinogenic compounds were identified in the combustion products, such as benzo(e)pyrene, benzo(a)anthracene and benzo(ghi)perylene.

MO455 Pt/Tk modelling of super-hydrophobic chemicals
W. Latisch, K. Goss, Helmholtz centre for environmental research - UFZ / Analytical Environmental Chemistry
It is a widespread opinion that super-hydrophobic chemicals are not taken up by fish [1–3]. But so far, we haven’t seen sufficient evidence why they should not cross membranes or aqueous boundary layers, although these processes might be kinetically slow. Super-hydrophobic chemicals are very challenging for controlled exposure experiments. Here we use the few existing data points of a fish feeding study with Dechlorane (log Kow (estimated) = 11.6) with our recently published Pt/Tk model, Tk-fish, to shed more light on this issue. We first validated the oral up-take pathway in our model and found that facilitated transport via albumin and bile micelles through the aqueous boundary layers must accounted for, for hydrophobic chemicals such as HCB in order to get correct results. Subsequent simulations with the super-hydrophobic chemical Dechlorane revealed that for an oral uptake route the difusive transport through aqueous boundary layers in the gastro-intestinal tract and in the blood is indeed the limiting process. Good agreement of the predicted model results with measured values indicates that there is no principal hindrance for the oral up-take of super-hydrophobic chemicals. The results also indicate that it would take roughly 2 years or more for a steady state to be established which is too long for an experimental exposure study. 1. Dyer SD, Bernhard MJ, Cowan-Ellsberry C, Perdu-Durand E, Demmerle S, Cravedi J-P. 2008. In vitro biotransformation of surfactants in fish. Part I: linear alkylbenzene sulfonate (C12-LAS) and alcohol ethoxylate (C13EO8). Chemosphere. 72:850—862. 2. Sakurutani Y, Noguchi Y, Kobayashi K, Yamada J, Nishihara T. 2008. Molecular size as a limiting characteristic for bioconcentration in fish. J. Environ. Biol. 29:89—92. 3. 2016. Guidance on Information Requirements and Chemical Safety Assessment, Chapter R.11: PBT/vPvB assessment Draft Version 3.0.European Chemicals Agency, Helsinki. https://echa.europa.eu/documents/10162/23047722/fr_csa_r11_pbt_peg_en.pdf/ddac903-0d4a-4995-93cc-3738162ba4e8

Migratory bird species at risk - the role of pesticides and other chemicals (P)

MO456 Main scientific gaps in knowledge of risk from pesticides to [migratory] wildlife globally, and potential contribution of WTIG to CMS questions
C.A. Bishop, Environment and Climate Change Canada / Wildlife Research Division

MO457 Main scientific gaps in knowledge of risk from rodenticides to [migratory] wildlife globally, and potential contribution of WTIG to CMS questions
P. Berny, VETAGRO-SUP / Toxicology

MO458 Main scientific gaps in knowledge of risk from Pb ammunition and shot to [migratory] wildlife globally, and potential contribution of WTIG to CMS questions
R. Cremin, Wildfowl & Wetlands Trust

MO459 Main scientific gaps on knowledge of risk from NSAIDs [migratory] wildlife globally, and potential contribution of WTIG to CMS questions
M. Taggart, University of the Highlands and Islands / Environmental Research Institute

MO460 Main scientific gaps in knowledge of deliberate poisoning to [migratory] wildlife globally
M. Odins, Independent Environmental Services Professional

Big data analysis in ecotoxicology: how to get new information out of existing data? (P)

TU001 Holistic evaluation of long-term field effect earthworm studies with the fungicide Boscalid
F. Shaab, BASF SE; J. Roembeck, S. Jaensch, ECT Oekotoxikologie GmbH; P. Kabou, BASF SE; S. Braeker, BASF France S.A.S.
In order to place a plant protection product on the market, the product and its active substances need to demonstrate an acceptable risk to earthworm communities. The current European risk assessment scheme follows a tiered approach using worst case environmental concentrations and endpoints from earthworm reproduction laboratory studies in tier 1. For the active ingredient Boscalid no risk to earthworms has been identified based on the chronic laboratory studies provided by BASF to EU registration authorities. However, for one of the formulated products containing Boscalid the tier 1 assessment did not allow to exclude a potential
long-term risk to earthworms in the field. Therefore, a comprehensive field study program was conducted in different crops and field sites in Germany between 2000 and 2010. The study program went beyond regulatory requirements and comprised 6 independent long-term field studies that ran up to five years and were accompanied by a comprehensive residue analysis program. The number of earthworm field data generated is - to our knowledge - one of the highest ever collected for one plant protection product. We evaluated the extensive data set under the assumption that these treatments put representativeness/comparability of examined earthworm communities, site- and soil properties of the different locations as criteria in the analysis. Based on these criteria a statistical assessments of representative and comparable earthworm communities in relation to the field exposure were conducted. The assessment revealed that – using data from representative and comparable study sites - there was no concentration related effect of a five-year use of the product regarding diversity and abundance of different earthworm communities.

TU002
Contextualising statistically significant differences observed in mesocosm studies using historical control data.
F. Joyce, Cambridge Environmental Assessments; H.S. Schuster, Cambridge Environmental Assessments (CEA) / Aquatic Ecotoxicology
Mesocosms (which aim to replicate communities residing in edge-of-field waterbodies) are used as part of the higher tier aquatic risk assessment for plant protection products (PPPs) in the EU registration process. When settingup mesocosm studies, care should be taken to standardise communities present in each replicate to reduce variability and maximise statistical power; indicated by minimum detectable differences (MDDs). However, being dynamic and complex systems, variability can often still occur between replicates, which can affect the reliability and interpretation of the results. Statistically significant differences can occur due to natural variability rather than biologically relevant effects, but documenting this to try and interpret the results of a single mesocosm study can be challenging. One option is to contextualise the experimental results from a single study using available historical control data; this is an approach often used for laboratory studies performed under standard conditions. It is, therefore, proposed that this approach can be extrapolated to mesocosm studies, given that they are also performed under standardised conditions as much as possible. Cambridge Environmental Assessments (CEA) have a wealth of control data from historical mesocosm studies, with samples collected in spring, summer and autumn, thus capturing the variability in population and community dynamics over multiple years and seasons. Here we present our review of this historical control data, and how this provides a baseline to aid interpretation of results from individual studies, allowing an assessment of biological relevance and thus the appropriateness of influencing the regulatory acceptable concentration (RAC). When integrated into the aquatic risk assessment, this will represent a realistic worst-case scenario.

TU003
Enhancing the utility of the ECOTOX knowledgebase via ontology-based semantic mapping.
The US Environmental Protection Agency’s Ecological Toxicology (ECOTOX) knowledgebase contains more than 30 years of reported single chemical toxicity effects data on aquatic and terrestrial organisms. Approximately 900,000 test results covering more than 11,000 chemicals and 12,000 species are available in ECOTOX. While the database is currently used by many sectors for a variety of purposes, a future goal is to allow for computational modeling of the data to identify novel adverse outcome pathways and networks, and assist in predicting species sensitivity. To accomplish these goals, the initial steps entailed: 1) validating the chemicals within ECOTOX 2) mapping species to NCBI taxids and 3) mapping all relevant ECOTOX codes to corresponding ontological terms so chemical effects data can be turned into computable phenotypic ontology classes. To semi-automate the code mapping, a Java-based lookup tool was developed using the ontology browser BioPortal (https://bioprotal.bioontology.org) as REST API to conduct bulk code mapping. This tool was designed to make use of BioPortal’s annotator and recommender functions so that all ontological class identifiers relevant to a particular ECOTOX term would be returned and specific ontologies recommended. Using this approach, the majority of the 2000+ ECOTOX codes were mapped to ontological class identifiers; some terms required multiple identifiers to properly describe them. Further, manual curation was needed during the rest cycle of a product is assessed via the USEtox multimedia fate model [3]. This model requires for each single chemical dozens of physico-chemical parameters as well as data of ecotoxicity to freshwater aquatic life and toxicity to human for cancer and non-cancer effects. For PEFLCA, those data are required for thousands of chemicals using the most up-to-date information [4,5]. The EU commission Joint Research Centre has obtained from the chemical agency (ECHA) for more than eight thousand chemicals all the physico-chemical properties (166'926 test results), ecotoxicity (242'729 test results) and human toxicity data (41'381 test results) available in the IUCLID 5.5 database (as of March 2017). The database has been used to calculate unique value for chemical properties

TU004
ECOTOX Knowledgebase: New tools for data visualization and database interoperability
C. Elonen, U.S. EPA/ORD/NHEERL; J. Olker, C. LaLone, U.S. EPA / Mid Continent Ecology Division; D.J. Hoff, U.S. EPA ORD / Mid Continent Ecology Division; S. Erickson, M. Skopinski, S. Casey, A. Pilli, K.A. Fay, CSRA, Inc. The ECOTOXicology knowledgebase (ECOTOX) is a comprehensive, curated database of toxicology data from single chemical exposure studies to terrestrial and aquatic organisms. The ECOTOX Knowledgebase provides risk assessors and researchers consistent information on toxic effects of chemical substances for use in deriving benchmarks and establishing criteria. ECOTOX has the capability to refine and filter data searches by 16 parameters (e.g. Species, Chemical, Effect, Control, Year, etc.) and customize output selections from over 100,000 fields. Study details such as species, taxonomic hierarchy, chemical purity, routes of exposure, and all calculated or statistically derived endpoints provided by the authors is encoded in discrete data fields for each test result. During the past 10+ years, ECOTOX has aligned the coding of the aquatic and terrestrial references by the addition of data fields, adapted search terminology to better focus literature searches, and updated search screens. To meet the data needs of 21st century toxicological assessments, new tools have been integrated into ECOTOX to improve data mining capabilities for end users such that environmental regulatory, the regulated industry, and researchers can more effectively and efficiently search and use existing toxic effects data. New data visualization and filtering options have been added to aid in data exploration. Efforts to enhance interoperability with other EPA databases have been employed to assist in efficiently accessing necessary data. These tools will be available in ECOTOX Knowledgebase version 5.0, to be released in FY18.

TU005
Edaphostat - A web application for automated and interactive meta-analysis of environmental data from the Edaphobase database.
J. Hausen, RWTH Aachen University; B. Scholz-Starke, M. Ross-Nickoll, RWTH Aachen University, Institute for Environmental Research / Institute for Environmental Research; R. Ottermanns, RWTH Aachen University / Institute for Environmental Research
Edaphobase is a database that summarizes toxicology data from single chemical exposure studies to terrestrial databases promises new insights in species occurrence and distribution, allowing e.g. the derivation of reference values and answering environmental questions on a larger scale. However, data from different studies are often very heterogeneous because the studies differ in scope, design, and parameters sampled. This makes meta-analysis a challenging task, as database users need to know how to select, combine and process this mixed origin-data. Automated analysis tools, which are customized for certain data warehouse applications, can be a solution to this problem. We present a web application called “Edaphostat”, which is part of the Edaphobase data warehouse (https://portal.edaphobase.org/). Edaphobase combines spatially explicit information on quantities of soil organisms, environmental parameters, and vegetation. The data in the warehouse are coming from different and diverse sources, ranging from large national-scale studies to o
biological complexity and representative of different trophic levels with the aim to establish a unique toxicity ranking. From the analysis of the results integration with TBI could be highlighted that to define the hazard associated with NPs is necessary to tailor the index parameters on specific NMs physico-chemical characterization. Moreover, to make the results more reliable, together with a larger number of tests, a longer testing time for some organisms and other endpoints (genotoxic and cytotoxic parameters) should be utilized.

TU007 Deriving physico-chemical input data for the USEtox model from the REACH database for thousands of chemicals using R-Studio program
F. Biganzoli, EU Commission Joint Research / Directorate D Sustainable Resources Bioconomy Unit; S. Proenca, EU Commission Joint Research; A. Gissi, European Chemicals Agency; E. Saouter, EU Commission JRC / Sustainable Assessment Unit
Product Environmental Footprint (PEF) and Organisational Environmental Footprint (OEFF) was a core part of the Commission Recommendation “on the use of common methods to measure and communicate the life cycle environmental performance of products and organisations” (2013/179/EU). The potential impact of chemicals emitted during the life cycle of a product is assessed via the USEtox multimedia fate model. This model requires for each single chemical dozens of physico-chemical parameters as well as data of ecotoxicity to freshwater aquatic life, terrestrial plants and animals, and ChEometric means with standard deviation - Comparison of the chemical toxicity based on the above calculations and chemical ranking based on GHS and CLP.

TU008 Toward a possible Toxicity Test Battery Integrated Index for Nanomaterials
M. Oliviero, University Parthenope; s. schiavo, ENEA CR; s. manzo, ENEA / SSPT-PROTER-BES
Nanotechnology is a rapidly expanding field of research continuously producing novel materials with nanoscale properties (nanomaterials, NMs), as result, it is inevitable that NMs will enter the aquatic environment. Usually, the ecotoxicological approach is generally based on a battery of bioassays with organisms belonging to various trophic levels, whose endpoints are not necessarily comparable for each species. While these endpoints, taken individually, can indicate the presence of a potentially deleterious effect, it is often difficult to combine these effects into an assessment of the overall status for the selected species. Therefore is necessary to integrate all results to evaluate the risk for NMs. To synthesize the results obtained with a battery of ecotoxicological tests, different approaches were selected as Toxicity test battery integrated index (TBI). This index has the advantage to put in evidence the differences between the samples. However there are still some open issues and TBI integration procedure needed to be modified according to testing sample or substance in order to represent the test sensitivity towards the matrices. In particular, when NMs are investigated also different physico-chemical behaviour and interaction with organisms should be taken into account. Therefore, the aim of this work is to study the suitability of TIB procedure for the NM to determine the needed modification for tailoring the data integration. In particular, we considered metal bearing nanoparticles (NPs) such as TiO2, SiO2 and ZnO and a battery of toxicity test with organisms of different toxicity indicators for thousands of chemicals to be used in the USEtox model. The poster presents the methodology applied for the selection of the aquatic toxicity data available in REACH, the set of criteria used to derive various level of quality data to meet the requirement to produced Effect Factors for as many chemicals as possible, and the various calculation procedure to derive final chemical effect factors. Correlation between acute and chronic toxicity for thousands of tests, for each chemical, group of chemicals, was established, as well as the feasibility to calculate effect values based on Species sensitivity distribution. The following final calculation have been performed for thousands of chemicals: - Acute and Chronic species geometric means with standard deviation and number of individual test available per species - Arithmetic average of all the log of the species geometric mean with standard deviation and count of species as well as count of SSD group for each chemical - Arithmetic average of all the log of the species geometric mean with standard deviation - Comparison of the chemical toxicity based on the above calculations and chemical ranking based on GHS and CLP.

TU009 Historical analysis of the use of plant protection products in apple orchards (1970-2014): Combining handwritten farmers records with electronic data
L. de Baan, Agroscope / Institute for Plant Production Sciences IPS; M. Mathis, J. Stieger; Agroscope; O. Daniel, Agroscope / Institute for Plant Production Sciences IPS
Plant protection products (PPPs) are used to protect crops against pests and diseases and ensure yields and quality of crops. Because they are biologically active, they can cause negative side effects on the environment or humans. Data-sets on the use of PPP for specific crops over a long time would allow to get a better knowledge on the potential emissions of PPP from the orchards and beyond agro-ecosystems. However, consistent long-term datasets are mostly lacking. In addition, historic farmers’ records are often only available in handwritten paper format. In Switzerland, data on PPP use in apple orchards has been voluntarily recorded by farmers since the 1950-ies, to evaluate farm economics. Up to the 1990-ies, data were only available in handwritten paper format, since 1997 they were collected electronically. In this study, we digitised the handwritten records and combined it with the electronic data. We first developed a concept, how the handwritten records can be entered into a database, which contains similar information as the electronic data. We collected data on farms (productivity), apple plantations (year of plantation, size, type, variety), and plant protection measures per plantation (product, dosage, date of application). In We also developed procedures to handle missing data and to detect errors in the data. A spray sequence in apple orchards over the period 1970-2014 was analysed, regarding the number of treatments per PPP category (e.g. fungicides), the total amount of active ingredients applied per season and PPP category and the ranking of chemical groups per PPP category. While the average number of treatments and the average amount of active ingredients remained within a similar range, major changes were detected in the composition of applied active ingredients. For example, in the 1970-ies and 80-ies, more than 75% of all insecticide treatments were organophosphates. After 1986, organophosphates were quickly replaced by carbamates and benzoylureas, and today only contribute to about 10% of all insecticide treatments. This study illustrates, that for a historical analysis of pollutants it is sometimes unavoidable to use manual handwritten data. In addition, the digital approach allows for a flexible and dynamic way to handle and to compare historic datasets.

TU010 Using long-term datasets to assess the impacts of neonicotinoids on farmland bird populations in the UK over the last 21 years
R. Lennon, The University of York / Environment; N. Isaac, NERC Centre for Ecology & Hydrology; R. Shore, Centre for Ecology & Hydrology (NERC); K. Arnold, University of York / Environment; W. Peach, Royal Society for the Protection of Birds; C. Brown, University of York / Environment Department. The abundance and diversity of farmland birds has been monitored by the majority of research regarding neonicotinoids (NNS) has been focused on pollinator species, however, little work has been done to investigate the potential long-term impacts of these pesticides on other taxa, such as farmland birds. Birds can be directly exposed to NNS via two main exposure routes: ingestion of NN-coated grain, or seedlings germinated from coated grain. With bird abundance data, NN usage records and UK cropping data, a poisson log-linear generalised mixed model was used to investigate whether there are any impacts of NN use on farmland bird populations over a period of 21 years. Specifically, the main objectives of this study were to 1) establish whether there is a relationship between species population growth and NN usage in the UK, 2) establish whether species traits, such as body weight and home range are correlated with any effect of NNS on species population growth, and 3) to ascertain whether the observational data (direct) was able to explain differences between the impacts of NNS on individual species population growth. A total of 54 bird species were modelled, for which the estimated effect of NNs on population growth were highly varied. Relationships between the estimated effects and species traits, including hypothesised risk to exposure will be reported.

TU011 Regression-based models reveal sources of pollutants in Norwegian marine sediments
G. Everaert, Flanders Marine Institute / Laboratory of Environmental Toxicology and Aquatic Ecology; A. Ruus, NIVA / NIVA; D. Hjernham, NIVA Norwegian Institute for Water Research; K. Borga, Department of Biosciences, University of Oslo / Department of Biosciences; N.W. Green, NIVA Norwegian Institute for Water Research / marine pollution; S. Boitsov, Institute of Marine Research; H. Jensen, Geological Survey of Norway; A. Poste, Norwegian Institute for Water
Research

We characterized spatial patterns of surface sediment concentrations of seven polychlorinated biphenyls (PCBs), seven polycyclic aromatic hydrocarbons (PAHs), three chlorinated pesticides and five metals in Norwegian waters and Skagerrak. In total, we analysed 5,036 concentrations of 22 chemical substances that were measured between 1986 and 2014 at 333 sampling sites by means of generalized additive models (GAMs). We found that GAMs with organic carbon content of the sediment and latitude as linear covariates explained ca. 75% of the variability of the contaminant sediment concentrations. For metals, a predominantly hotspot-driven spatial pattern was found, i.e. we identified historical pollution hotspots (e.g. Sørked in western Norway) for mercury, zinc, cadmium and lead. Highest concentrations of PAHs and PCBs were found close to densely populated and industrialised regions, i.e. in the North Sea and in the Kattegat and Skagerrak. The spatial pattern of the PCBs suggests the secondary and diffuse atmospheric nature of their sources. Atmospheric inputs are the main sources of pollution for most organic chemicals considered, but north of the Arctic circle, we found that concentrations of PAHs increased from south to north most likely related to a combination of coal-eroding bedrock and the biological pump. The knowledge acquired in the present research is essential for developing effective remediation strategies that are consistent with international conventions on pollution control.

TU012
Application of a ‘weight-of-evidence’ model for assessing sediment quality and associated hazard with offshore gas platforms discharging produced water

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A multidisciplinary weight of evidence (WOE) study was carried out, integrating different lines of evidence (LOE) as sediment grain size analysis and bioaccumulation on native mussels. In this study, a chemically characterized by sampling of twenty-four stations at increasing distance from the platform/discharge, and in particular four stations, located at 0, 25, 50 and 100 m along the main local current, also for ecotoxicity. Different inorganic and organic contaminants were analyzed in sediment. To assess the effects of pollutants at different levels of trophic web, results of a bioassays battery composed by the Vibrio fischeri, Dunaliella tertiolecta, Skagerrak. In total, we analysed 5,036 concentrations of 22 chemical substances (PCBs), seven polycyclic aromatized additive models (GAMs). We found that GAMs with organic carbon content of the sediment and latitude as linear covariates explained ca. 75% of the variability of the contaminant sediment concentrations. For metals, a predominantly hotspot-driven spatial pattern was found, i.e. we identified historical pollution hotspots (e.g. Sørked in western Norway) for mercury, zinc, cadmium and lead. Highest concentrations of PAHs and PCBs were found close to densely populated and industrialised regions, i.e. in the North Sea and in the Kattegat and Skagerrak. The spatial pattern of the PCBs suggests the secondary and diffuse atmospheric nature of their sources. Atmospheric inputs are the main sources of pollution for most organic chemicals considered, but north of the Arctic circle, we found that concentrations of PAHs increased from south to north most likely related to a combination of coal-eroding bedrock and the biological pump. The knowledge acquired in the present research is essential for developing effective remediation strategies that are consistent with international conventions on pollution control.

TU013
Using biomarkers in a multispecies approach to relate organochlorine exposure and biological effects

V. Weyneux, North-West University - School of Biological Sciences / School of Biological Sciences, University of Venda / Zoology; R. Gerber, North-West University / Unit for Environmental Sciences and Management; N. Smit, North-West University / Environmental Sciences and Management

Due to their persistence and global distribution the use of organochlorine pesticides (OCPs) have been banned in most countries around the world. However, in a number of countries, such as South Africa, the use of DDT as a vector control agent is still allowed. This practice is not without controversy and reports on ecological and human health effects are increasing. The Phongolo River floodplain in north-eastern South Africa is a high risk malaria area where DDT is used as vector control agent through indoor residual spraying (IRS). This region is also regarded as a biodiversity hotspot in southern Africa and concern has been raised regarding the risk posed as a result of the long term use of DDT. Over the past seven year’s studies have been undertaken to determine the degree of DDT exposure in the aquatic ecosystem through analyzing DDT and other OCP bioaccumulation in a number of different aquatic species. Concomitant biomarker analyses were undertaken to determine the biological effect of the DDT exposure. In this poster we collated and integrated the exposure (DDT and HCH bioaccumulation) and effect (biomarker) data of the different studies to test the hypothesis that increased DDT exposure will elicit similar biological responses across species. Bioaccumulation of DDT (and its metabolites) and HCHs were measured before and after IRS application periods in two decapoda, six fish and two amphibian species. Biomarkers of exposure (cytochrome P450 and acetylcholine esterase) and effect (catelase, superoxide dismutase, malondialdehyde, protein carbonyl, and cellular energy allocation) were analysed in the same organisms. Using principal component analysis and discriminant functional analysis the exposure and effect data were integrated to elucidate the responses of aquatic biota to OCP exposure. Although higher trophic level organisms (i.e. tigerfish - Hydrocynus vittatus and Muller’s clawed frog - Xenopus laevis) displayed a significant increase in DDT bioaccumulation there were no distinct biomarker responses evident. When exposure data of another banned OCP, γ-Hexachlorocyclohexane, were included in the analysis, significant relationships with cytochrome P450 and lipid energy reserves were obtained. The result therefore indicated that biological responses were not related to DDT but rather to HCH exposure.

Microbial community ecotoxicology in environmental risk assessment and ecosystem monitoring (P)

M. Simonin, Duke University / Biology; K.A. Voss, Regis University; B.A. Hassett, J.D. Roca, S. Wang, Duke University / Biology department; C.R. Violin, University of North Carolina at Chapel Hill / Biology department; E.S. Bernhardt, Duke University / Biology department.

The advent of high throughput sequencing enabled microbial ecotoxicologists to better characterize the impact of stressors and especially pollution on microbial community structure. However, the overwhelming amount of information generated by sequencing and the high diversity of microorganisms led us to focus our analyses mainly at the community or phylum levels, ignoring all the key ecological knowledge potentially gained at the population level. In this study, we tried to move beyond bulk communities by using a high-throughput sequencing platform. We characterized spatial patterns of surface sediment concentrations of seven inorganic and organic contaminants were analyzed in sediment. To assess the effects of pollutants at different levels of trophic web, results of a bioassays battery composed by the Vibrio fischeri, Dunaliella tertiolecta, Skagerrak. In total, we analysed 5,036 concentrations of 22 chemical substances (PCBs), seven polycyclic aromatized additive models (GAMs). We found that GAMs with organic carbon content of the sediment and latitude as linear covariates explained ca. 75% of the variability of the contaminant sediment concentrations. For metals, a predominantly hotspot-driven spatial pattern was found, i.e. we identified historical pollution hotspots (e.g. Sørked in western Norway) for mercury, zinc, cadmium and lead. Highest concentrations of PAHs and PCBs were found close to densely populated and industrialised regions, i.e. in the North Sea and in the Kattegat and Skagerrak. The spatial pattern of the PCBs suggests the secondary and diffuse atmospheric nature of their sources. Atmospheric inputs are the main sources of pollution for most organic chemicals considered, but north of the Arctic circle, we found that concentrations of PAHs increased from south to north most likely related to a combination of coal-eroding bedrock and the biological pump. The knowledge acquired in the present research is essential for developing effective remediation strategies that are consistent with international conventions on pollution control.
during one month was exposed in channels at 6 increasing concentrations of diuron: 0, 1, 5, 10, 25 and 50 µg.L\(^{-1}\) for two hours, with a flow velocity of 2 cm.s\(^{-1}\). Then, Langmuir isotherm equation (Praus et al. 2007) was fitted to the bioaccumulation data. During the determination of the isotherm, a plateau was reached over 5 µg.L\(^{-1}\) of diuron in the water. This suggested that all absorption sites were saturated, and then diuron concentration in the biofilm became independent of diuron concentration in the water. The fitting of a Langmuir isotherm allowed to estimate a maximum uptake of 1439.073 µg.g\(^{-1}\) and an equilibrium constant of 0.378. Photosynthesis inhibition was correlated \((R^2=0.75)\) to diuron concentration in the water. The data did not clearly highlight a relationship between bioaccumulation and photosynthesis inhibition. This study establishes that diuron bioaccumulation in biofilm is nonlinear, and allows to calculate the equilibrium and maximal capacity of the biofilm regarding diuron uptake. These two constants can be used to further prediction of diuron bioaccumulated in biofilm from concentration in the water. The innovative coupling of toxicokinetic and toxicodynamic approaches would provide original information about absorption behaviour and impact in periphytic microorganisms.

TU016

New insights into the biotransformation of sulfurlamide: role of ammonia oxidizing bacteria and community shifts

T. Yin, National University of Singapore / Civil and Environmental Engineering; Y. Yang, S. Te, National University of Singapore; K. Gin, National University of Singapore / Civil & Environmental Engineering

Emerging organic contaminants (EOCs), such as perfluoralkyl and polyfluoroalkyl substances (PFASs), are ubiquitously detected in the environment and have raised increasing concerns due to their adverse effects on ecosystems and humans. N-ethyl perfluorooctane sulfonamide (N-EtFOSA), belonging to PFASs, is used as the active ingredient in the pesticide. Sulfurlamide, which is particularly important in the control of leaf-cutting ants in some developing countries. Previous studies have investigated the bioaccumulation in different biotopes and the biotransformation in activated sludge, marine sediments and soil. However, little information is available on the contributions of different microbes to the biotransformation of N-EtFOSA. This study used Allotrychium (ATU), an inhibitor of ammonia monooxygenase (AMO), to investigate the relative contributions of ammonia oxidizing bacteria (AOB) and other members to N-EtFOSA biotransformation and find potential N-EtFOSA degraders by analysing the microbial community shifts. In the reactors with ATU addition, N-EtFOSA was degraded faster with an apparent half-life of 1.3 days, which indicated that ATU had actually enhanced the biotransformation of N-EtFOSA. This implied that AMO was probably not involved in the biotransformation of N-EtFOSA, and thus the inhibition of AMO by ATU had no adverse effect on its biotransformation. ATU-treated sample was more diverse with a Shannon index of 4.04 while that of the ATU-untreated sample was 2.43. The abundance of Candidatus Protochlamydia increased significantly in the ATU-treated sample, which suggested that this genera could be a potential degrader for N-EtFOSA. Future work needs to determine the genes involved in the biotransformation process using metagenomics and metatranscriptomics.

TU017

How can three herbicides impact the fatty acids of the freshwater diatom Gomphonema gracile ?

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Fatty acids are essential elements for the structure of biological membranes and for the storage of metabolic energy. They are used as a source of energy by metabolism at each trophic level, making fatty acids biochemically and physiologically important compounds (Neves et al. 2015). In the trophic chain, many fatty acids are only synthesized by microalgae and bacteria before being transferred via herbivorous invertebrates to fish and ultimately to humans (Arts et al. 2001). For example, highly unsaturated fatty acids (HUFAs) such as eicosapentaenoic acid (EPA; C20:5n3), can not be synthesized de novo or in insufficient proportions by animals, and need to be preformed in plants that is why they are used to study trophic interactions in food chains. Generally, microalgae with a high proportion of EPA, such as diatoms, are an excellent source of food for animals but the concentrations of these different fatty acids can vary according to the stage of growth of the organism and according to different environmental parameters including pesticide exposure (Brett et al. 2006, Robert et al. 2007, Burns et al. 2011, Fink and Fu et al. 2016). Moreover, for several years, the intensive use of pesticides has caused many problems to the environment, making pesticides major pollutants of aquatic ecosystems (Aydinalp and Porca 2004). The aim of this study is to investigate the impact of 3 pesticides on diatom’s fatty acids. To address this issue, a model freshwater diatom (Gomphonema gracile) was exposed to three herbicides, with three different cellular targets, at environmentally relevant and higher concentrations (diuron and S-metolachlor, C1=1 µg.L\(^{-1}\) and C2=10 µg.L\(^{-1}\); glyphosate, C1=5 µg.L\(^{-1}\) and C2=50 µg.L\(^{-1}\)). After a 1-week exposure, fatty acid compositions of diatoms were determined by gas chromatography. In comparison with control samples the percentage of 1) polyunsaturated fatty acids (PUFA) decreased with S-metolachlor contamination (C2); 2) saturated fatty acid (SFA) and monounsaturated (MUF A) decreased with diuron and glyphosate exposure (C2). The decrease of PUFA is a direct impact and can be explained by the mode of action of S-metolachlor which inhibits elongases. Concerning diuron and glyphosate, the decrease of SFA and MUF A can reflect an indirect effect, which can be explained by the mode of action of these two pesticides which respectively blocks electron transfer in photosynthesis, and inhibits the synthesis of aromatic aminocids.

TU018

Effects of Nickel on cell cycle progression, growth and antioxidant enzymes of green algae C. reinhardii

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Freshwater ecosystems received industrial and domestic sewage discharged and natural chemical compounds as a result of anthropogenic activities. Heavy metals released in the environment have increased over the last decades causing environmental and human health problems worldwide. The known biological adverse effect of metals include growth disorders, disturbances of membrane functions and pigments synthesis pathways, induction of oxidative stress, mutagenic effects, among others. Among aquatic organism, microalgae have an important role in aquatic system as they are a key component of food chains. So that, it is crucial to has early assessment tools to evaluate effects of metals at the cellular level. In the present study effects of Nickel was evaluated on cell cycle progression, growth and antioxidant enzymes kinetic of the green algae C. reinhardii. Synchronized cultures of this multiple fission dividing algae were used for the study. Aliquot from growing cultures were taken hourly during 36 hours. The attainment of commitment points (CP) was evaluated by transferring hourly aliquot into aerated tubes at 30 °C in the dark. Analysis of cellular division, nuclear division (DAPI stain) changes in cell size, were performed. The proportion of mother cells and daughter cells were assessed at the end of the cell cycle. Toxicity of metal was assessed by algal growth inhibition test, estimating toxicity endpoints, growth rates, protein, antioxidant enzymes activities of catalase, guaiacol peroxidase, ascorbate peroxidase, glutathione reductase and concentration of chlorophyll a, chlorophyll b and carotenoids at the end of 96 h of exposition. Nickel provoked a block of cell cycle at the highest concentration tested. At lower concentrations, cell cycle progression was observed with different pattern of attained CP, depending the exposure concentration. Antioxidant enzyme activities were inhibited at concentration above 0.05 and 1 mg/L. The effects of metal on pigment concentration was less evident than the effects on growth rates, indicating a lower sensitivity of these parameters. Nickel provoked severe damage on algal cell division cycle, the cycle progression was observed. At the detection of the amount of applied fertilizer. Zinc oxide NPs have been largely used nanofertilizers their spreading to crops seems to increase their growth and yield. This increasing use could lead to their introduction in the environment occurring also via unintentionally pathways. Actually, some studies reported ZnO NPs negatively affect soil microbial activities and consequently the biogeochemical cycles. These effects could be evidenced by assessing metabolic profiles of culturable, aerobic, heterotrophic microorganisms (Biolog). BiologistEcopele was successfully used to detect short and long-term changes of functional diversity of soil microbial communities. This method was based on the determination of the oxide profiles related to several different carbon sources. This study aims to investigate the changes in the metabolic profile of culturable soil microorganisms along decimal dilution in response to the exposure for one month to two different type of fertilizers (F1 and F2). The fertilizers were added with ZnO Bulk, ZnO NPs and ionic zinc (ZnSO\(_4\)) at 230 mg Zn/kg. Then, the fertilizers with Zn compounds were added to the farm soil. After 15 days of soil exposure to fertilizers with ZnO Bulk, NPs and ZnSO\(_4\), the eluates were obtained. Different eluates were tested upon, Pseudokirchneriella subcapitata (6.25%, 12.5%, 25% and 50%), Lepidium sativum. At time 0, 15, 30 days fresh soil samples were assessed by using the BiologistEcopele approach. The occurrence of the microbial oxidation of each BiologistEcopeleTM M source was calculated as probability ‘p’ on a binomial scale of data in order to identify the treatments able to preserve the highest possible oxidizing ability of C substrates and those negatively affecting it. ZnO-nanofertilizers were more toxic than fertilizers with ZnO in bulk form, for algal growth, instead for plants, the effects of “ZnO-nanofertilizers” depended on the fertilizer type: only F1 + ZnO NPs resulted more stimulating than F1 + ZnO Bulk. Preliminary Biolog results seemed to highlight that the microbial community
was mainly affected by ZnO NPs. The integration of the classical ecotoxicology with BiogeoEcoTale approach could represent a good strategy to establish the environmental risk related to the use of nanofertilizers. Keywords: microbial community, ecotoxicology, nanofertilizers

TU0/20
Environmental factors-regulated disease dynamics of tilapia lake virus (TilLV) transmission in farmed tilapia ponds

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BACKGROUND: Outbreaks of tilapia lake virus (TilLV) have caused substantial mortalities of farmed tilapia, posing a significant threat to worldwide tilapia industry. Environmental factors controlling TilLV disease dynamics should be clearly elucidated to prevent the potential economic impacts on aquaculture.

OBJECTIVE: The main objective of this study was to make the TilLV disease dynamics by constructing an epidemiological model to implicate aquaculture management among farmed tilapia ponds.

METHODS: The mortality of Nile tilapia infected by intraperitoneal (I.P.) injection with different TilLV dosage were fitted by two-parameter Hill model to estimate median lethal dose (LD50). To explore TilLV highly artificial environmental conditions. Sacrificing some of the susceptible-infectious-mortality (SIM) model was applied to describe cumulative mortality data to estimate mortality rate (α), transmission rate (β), and basic reproductive number (R0) for Nile tilapia pose by TilLV under treatment of cohabitation.

RESULTS: In toxicity assessment, LD50 estimate of Nile tilapia infected by I.P. injection with different TilLV dosage was 5.71±2.7 TCID50 mL-1. The mortality of Nile tilapia was not inhibited by TiO2 nanoparticles explained CuO toxicity, TiO2 nanoparticles could provide valuable information using different model organisms, experimental endpoints and conditions could be applied to develop a rapid method for toxicity detection. The observed discrepancies in toxicity indicate that the act of autochthonous microbial communities able to break down the more chlorinated compound to lesser chlorinated ethenes which need to be detoxified as well. Bacteria able to metabolize such toxic substances are indeed well known as well as many of their metabolic pathways, but still an efficient and complete detoxification process is hard to achieve. The understanding of the microbial activity underpinning the whole process is crucial especially during a bioremediation process where microbes are stimulated through the amendment of nutrients in order to obtain the complete detoxification. The huge impact of metagenomics, and other molecular biology techniques for the comprehension of microbial composition and activities in different environments, is helping to shed light for the comprehension of the critical apparatus behind the detoxification process but we are still at the beginning. During the present work, two microbial populations inhabiting a chlorinated solvent polluted groundwater, with and without nutrient amendment, have been analyzed after whole genomic DNA extraction and sequencing. These observations suggest that the microbial communities’ structure and/or functional composition differ between the two tested substrates as well as among different environmental factors such as temperature and aquaculture density. Results of toxicity assessment and disease epidemics could provide insights into aquaculture management of TilLV disease by controlling potential factors in tilapia ponds.

Keywords: Tilapia lake virus; Toxicity assessment; Susceptible-infectious-mortality model; Aquaculture management

TU0/21
Natural organic matter alleviates TiO2 and CuO nanoparticle toxicity in four algal species

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Although the knowledge concerning synthetic metal nanoparticle (NP) effects on aquatic organisms has improved during the last decade, more research has been conducted on TilLV highly artificial environmental conditions. Sacrificing some of the experimental reproducibility to obtain more environmentally relevant data, we studied the 72-hour effects of uncoated CuO (CuSO4 as tonic control) and TiO2 NPs on two endpoints, biomass production and photosynthetic maximum quantum yield (Fv/Fm), in nutrient-adjusted natural water (ANW) and the OECD201 standard medium, using four freshwater species from three major algal groups: green algae (Ulva lactuca), blue-green algae (Synechocystis sp), and cyanobacteria (Synechocystis sp). Metal toxicity on both parameters at 72 h was reduced in ANW in all algal species except the cyanobacterium, presumably because of natural organic matter (NOM) binding to the NPs and solubilized ions. The biofilm-forming diatom was most resistant to NPs when incubated in ANW, whereas both the diatom and the cyanobacterium were not inhibited by TiO2 at concentrations up to 100 mg/L throughout. TiO2 significantly inhibited biomass production of both green algae in the standard medium (EC50: 14.1-31.4 mg/L), but only R. subcapitata was inhibited in ANW (EC50: 31 mg/L). TiO2 NPs did not significantly inhibit Fv/Fm of any species in either medium up to 100 mg/L, indicating a lack of toxic effect on the photosynthetic apparatus. The sensitivity to CuO remained at a similar level in the standard OECD medium (biomass based EC50: 0.2-0.9 mg/L) for all species, but differed over orders of magnitude in ANW (EC50: 0.3-16 mg/L). The cyanobacterium, that has the smallest cell of the four tested species, was consistently most susceptible to Cu toxicity. While shedding of Cu ions from particles explained Cu toxicity, TiO2 effects were at least in part due to observed cell/organism heterogametization. Overall, Fv/Fm was less a sensitive toxicity endpoint than biomass, but the two parameters were strongly correlated (Spearman’s p=0.6-0.9) when toxicity was evident, again proving Fv/Fm as a rapid method for toxicity detection. The observed discrepancies in toxicity indicate that using different model organisms, experimental endpoints and conditions could provide valuable information about the behavior of emerging contaminants in the environment, thus improving the quality of risk assessment. Research was funded by IUT23-5.
Environment Health; L. Mancini, Italian Institute of Health ISS; C. Mancini, AECOM / Environment Health; M. Delledonne, M. Rossato, University of Verona; F. Fatone, Università Politecnica delle Marche; A. Farinelli, J. Blum, D. Bloisi, University of Verona; A. Cordioli, Azienda Garesdane Servizi S.P.A. Peschiera del Garda; P. Varotto, Azienda Garesdane S.P.A. Peschiera del Garda; A. Titonnel, Technical S.P.A. Milano; D. Calisi, Algorithmica S.r.l. Roma; F. Giannone, Algorithmica S.r.l.; R. Allahabadi, Boku University; A. Parsons, L. Parsons, Denbiotest Ltd.d.; T. Runnalls, Brunel University / IFE; G.E. Brighty, Environmental Sustainability Associates limited; T. Licha, Göttingen University; S. Malamis, Athens Technical University; T. Knutz, Go-Sys; A. Merkoci, ICREA - The European Project Horizon 2020 INTCATCH (Development and application of Novel, Integrated Tools for monitoring and managing Catchments) has the main goal to recommend and deliver a new approach and methodology for the monitoring and assessment of surface waterbodies in Europe. The tools foreseen by INTCATCH include sensors for the detection of heavy metals, nutrients, pH, temperature, pesticides, Escherichia coli, some of them are mounted on aquatic drones. An innovative tool of Intactch is the portable sequencing laboratory/tool for metagenomic analysis that is performed in different surface waterbodies (e.g. Garda lake) selected through an analysis of the pressures. Bacteria are collected after concentration of water samples that are filtered by an automatic system applied on the aquatic drones. Using portable devices, the genomic tool allows the DNA meta-barcoding on-site, where samples are collected, and deliver the full bacterial composition of the water analyzed, including pathogens. The validation of the metagenomic results is performed with the use of traditional microbiological analysis of raw water. The metagenomic data can support the traditional monitoring tools used in sediments, although contamination because changes of the bacterial community can reflect variations in water quality, and be linked for example to the presence of unknown chemical substances or mixtures. In addition, given its capability to detect pathogens, such portable genomic tool can provide a valuable rapid readout in case of emergencies, that are increasing in frequency due to the effects of climate changes, such as flooding. Similarly, by using the metagenomics data, linked to the informations of the other tools, can be also used for the identification of pollution sources because the proportions of the bacteria groups vary in relation to several stressors (agricultural, urban, living stock, etc.).

TU025 Tolerance of sediment-microbial communities to copper indicates lake contamination A. Tili, Eawag / Department of Environmental Toxicology; C. Bonineau, Isetra Lyon; A. Dabin, Isetre Lyon-Villeurbanne / UR MALY; E. Lyautey, Universite Savoie Mont Blanc; B. Ferrari, Centre Ecotox EAWAGEPFL; S. Pesce, Isetre Lyon-Villeurbanne / Microbial ecology of anthropised river systems. In lakes, and fresh waters in general, sediments play a crucial role in biogeochemical cycles, and are often at the basis of the food-web. Heavy metals such as copper, zinc or silver can accumulate in lake sediments, which represent a risk for bentthic microorganisms, with potential negative effects on the functioning of the whole ecosystem. Therefore, the goal of this field survey was to examine the impact of copper on microbial communities in sediments, describing copper contamination by heavy metals in lake Geneva. Sediments were sampled with an Ekman grab sampler at four sites, located around the lake and close to the shore. Samples were analysed for total metal concentrations, organic matter content, abundance of metal-resistance genes (e.g copA and cusA), bacterial biomass, and bacterial community composition. Moreover, tolerance of microbial communities to copper was evaluated using a transcriptional response of genes (TARGOT) concept by measuring the inhibition of bacterial secondary production and the extracellular enzyme β-glucosidase activity. Results showed a clear gradient of copper contamination, ranging from 30 to 350 mg.kg⁻¹ DM sediment. The results also demonstrated that chronic in situ exposure to copper induced a decrease of bacterial biomass and a structural shift in the community composition. Interestingly, tolerance measurements to copper were strongly and positively correlated to the copper concentrations in the sediments. Overall, our findings support the fact that microbial communities in sediments are a good indicator for metal contamination in aquatic ecosystems, and the suitability of microbial tolerance measurements to pinpoint specific effects of metals at the community level.

TU026 Current challenges and perspectives in aquatic and soil microbial community ecotoxicology K.K. Brandl, University of Copenhagen / Department of Plant and Environmental Sciences; M. Schmidt-Jürgens, UFZ - Helmholtz Centre for Environmental Research; A. Tili, Eawag / Department of Environmental Toxicology Assessing the impacts of chemicals still largely relies on approaches that are based on assays in simplified laboratory settings at the single-species or sub-species level. Yet, it becomes increasingly recognised that to draw conclusions on potential effects on ecosystems, it is essential to consider higher levels of ecological organisation, and more intricate risks on ecosystem structure and function. Microbial communities provide a large range of ecosystem services such as primary and secondary production, nutrient recycling, pollutant degradation, and are sources of biochemicals. Microorganisms are also primary targets for chemicals, which can lead to structural and functional alterations of microbial communities, with potential negative consequences for ecosystem functioning and environmental selection of antimicrobial resistance. Hence, a microbial community-level perspective in ecotoxicology is more important than ever. In this presentation we will first provide an overview on the current status of microbial community ecotoxicology research, in aquatic and terrestrial ecosystems, and to which extent this field is considered in environmental risk assessments. After that, we will describe the challenges within both aquatic and terrestrial microbial community ecotoxicology. Finally we will discuss future research directions in microbial community ecotoxicology to accurately assess and predict impacts of chemicals on ecosystems, and to develop specific response indicators of chemical exposure and effects.

TU027 Hydrodynamic conditions alter the tolerance of biofilm communities towards chemical stress B. Holzhott, Helmholtz Centre for Environmental Research - UFZ / Department of Bioanalytical Ecotoxicology; F. Larra, Helmholtz Centre for Environmental Research - UFZ GmbH; S. Lips, Helmholtz Centre for Environmental Research UFZ / Department of Bioanalytical Ecotoxicology; C. Anlanger, U. Risse-Buhli, M. Weitere, Helmholtz Centre for Environmental Research UFZ / Department of River Ecology; M. Schmitt-Janousek, UFZ - Helmholtz Centre for Environmental Research / Department of Bioanalytical Ecotoxicology Biofilms in rivers are complex communities built of bacteria, fungi, algae and protozoa embedded in a matrix of extracellular polymeric substances (EPS). They are important hotspots for biogeochemical processes in aquatic systems. A variety of stressors can potentially affect the structure and function of biofilms. Therefore, their tolerance to one stressor may be influenced by former exposures to another stressor. Community composition and physical structure is influenced by EPS hydrodynamics. Even though the influence of EPS hydrodynamics are reported to decrease with higher mean flow velocity and turbulence, the cell-to-EPS ratio increased. As the EPS content of a biofilm may influence the bioavailability of toxicants, differences in community tolerance towards herbicides are expected for biofilms grown under variable flow conditions. Still, the interactive effects of hydrodynamic growth conditions and herbicide tolerance are lacking.Using an artificial flow-through channel and water from the River Selke (Elbe catchment, Germany), we created heterogeneous flow regimes and related biofilm community structure and function to different mean flow velocities and values of turbulent kinetic energy. Taking the biofilms grown under such controlled hydraulic conditions, herbicide tolerance towards prometryn was tested according to the OECD test method. Focusing on the phototrophic part of the biofilm communities, we 1) investigated the algal structure, function and herbicide tolerance under different near-bed turbulences (diatom composition, photosynthesis) and 2) assessed the role of EPS in stressor interactions. The relevance of EPS content in combined stressor interactions was confirmed by using artificial EPS and algal cultures.

TU028 Does fungicide exposure alter interspecific relationships of aquatic fungi during leaf-decomposition? - A case study using species-specific qPCR assays N. Roedig, University of Koblenz-Landau; P.T. Baudy, University of Koblenz-Landau / Institute of Environmental Sciences; J.P. Zubrod, D. Englert, University of Koblenz-Landau / Institute for Environmental Sciences; M. Kowalshk, University Koblenz-Landau / Institute for Environmental Sciences; C. Baschien, Leibniz Institute DSMZ - German Collection of Microorganisms and Cell Cultures; R. Schulz, University of Koblenz-Landau / Institute for Environmental Sciences; M. Bendschuh, Swedish University of Agricultural Sciences / Department of Aquatic Sciences and Assessment Aquatic hyphomycetes, a polyphyletic group of freshwater fungi, are considered key players in leaf litter breakdown, an important ecosystem function in aquatic systems. Structural implications of anthropogenic stressors in aquatic hyphomycete communities have mainly been analysed using spore morphology, which does not allow assessing direct inferences on species-specific abundance and performance under stress. Therefore, we performed a microcosm experiment in which we related the microbial structure, function and herbicide tolerance under different near-bed turbulences (diatom composition, photosynthesis) and 2) assessed the role of EPS in stressor interactions. The relevance of EPS content in combined stressor interactions was confirmed by using artificial EPS and algal cultures.

Aquatic hyphomycetes, a polyphyletic group of freshwater fungi, are considered key players in leaf litter breakdown, an important ecosystem function in aquatic systems. Structural implications of anthropogenic stressors in aquatic hyphomycete communities have mainly been analysed using spore morphology, which does not allow assessing direct inferences on species-specific abundance and performance under stress. Therefore, we performed a microcosm experiment in which we related the microbial structure, function and herbicide tolerance under different near-bed turbulences (diatom composition, photosynthesis) and 2) assessed the role of EPS in stressor interactions. The relevance of EPS content in combined stressor interactions was confirmed by using artificial EPS and algal cultures.
resulted in an up to 99% reduced abundance of the inferior species. Species interactions were largely unaffected by fungicide exposure as dominant species where generally those identified as tolerant towards fungicide exposure. However, qPCR results revealed that one of the two sensitive species exhibited a significantly increased DNA yield in presence of the other one at field-relevant fungicide concentrations (5 µg/L). Species-specific qPCR assays proved to be a valuable tool for assessing ecotoxicological effects on well-defined ecological interactions within aquatic hyphomycete communities. In the future, this technique might become an asset in aquatic risk assessment and environmental stress monitoring.

TU029
Cyanobacterial Bloom in the Lake Varese: Characterisation of Microbial Communities by Metagenomics analysis
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The increasing anthropogenic eutrophication and climate changes are contributing to the intense proliferation of cyanobacteria in waterbodies so causing a phenomenon known as bloom which may compromise the quality of drinking and recreational water. The dynamics of bloom events are not yet fully understood, however it is scientifically accepted that external factors such as water temperature, nutrient concentrations and light intensity, may influence the potential of a bloom. Our study focuses on the relationship between environmental factors and the composition of the microbial community of the lake Varese (Italy) for a period of several weeks before and after the bloom event. Sampling campaigns were performed on a weekly basis. Water samples were collected from the depth region of the lake (0.5 to 3 m) and subsequent water column depths, 0.5 m from surface depth (0.5 m) and 2.5 times the Secchi disk depth measured in situ on sampling day (2.5x SECCCHI). The samples were characterised for their chlorophyll a content, nutrients, cyanatoxins and genomic DNA was extracted for metagenomics. Purified DNA samples were subjected to 16S sequencing (variable region V3-V4) and for shotgun analysis. All 16S samples were MiSeq sequenced as 2x250bp paired reads, the corresponding shotgun samples as 100bp paired reads. Shotgun analysis was performed for sample collected from 31/8/2016 until 5/10/2016 and only for EPI and 2.5x SECCCHI. The results showed that a peak of cyanobacteria was observed around 14.9/21.9 in the EPI (E) samples consistent with the high observed concentration of chlorophyll a. The lowest abundance of the cyanobacteria was in the SECCCHI (C) samples. Community composition of the overall cyanobacterial composition was also observed for proteobacteria and actinobacteria. Our result suggests that the major differences in bacterial community composition during the bloom are concentrated in the SECCCHI depth region while composition of the EPI zone is more or less constant. Cyanobacteria were found highly abundant in Lake Varese and are therefore likely responsible for the bloom. This hypothesis is also supported by the cyanotoxin data although complex and varying 18S metagenomes would be recommended in order to discard a possible contribution of phototrophic eukaryotes.

TU030
Following copper bioaccumulation and internalization during freshwater biofilm development using stable Cu isotope
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In small streams, microbial communities form river biofilms attached to solid substrates by producing extracellular polymeric substances (EPS). This matrix may act as a protective layer by limiting cellular contact with surface water contaminants. Thus, several studies have suggested that during biofilm growth, biofilm and EPS matrix thickness could limit cellular bioaccumulation. To test this hypothesis, 38 cu biofilm cultures derived from the upper reaches of the Orgeval river were cultured under a Cu stress environment by growing the biofilm on glass slides in water of concentrations of 0.06 uM and higher. PICT measurements confirmed that copper concentrations in a mature biofilm at a given time reflect the last period of exposure. In addition, analysis of the cell fraction showed that in the surface, copper was very low, a significant difference of isotopic ratios between the EPS fraction (0.25) and the cell fraction (0.35) confirms the potential protective function of the EPS matrix.

TU031
Zirconium impact on freshwater periphytic communities
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The growing world demand for metals increases metallic element mobilization in aquatic systems. Although the effect of metals on freshwater ecosystems is well documented, studies on the impacts of tetravalent metals are very scarce. Zirconium (Zr) is a tetravalent non-radioactive element for which the global demand has been increasing in the last decades. Benthic microorganisms community (periphyton) have shown good potential as a biomonitoring tool to assess metal exposure of aquatic organisms. In this work, the effect of Zr on periphyton biodiversity and biochemistry was investigated to apply this tool to zirconium and other tetravalent metals contamination assessment and to better understand their potential impacts on aquatic ecosystems. Glass slides were immerged in a pond in Cestas (near Bordeaux, France) for one month to be colonised by periphyton. They were distributed in 3 aquarium containing a synthetic culture medium (Dauta, 1982) and effective concentrations of 0.2 ± 0.1 nM (CO), 0.5 ± 0.3 nM (C1) or 2.9 ± 0.3 nM (C2) of Zr (neq). One slide per section was sampled after 1, 2 and 4 weeks of exposure. DNA samples were subjected to 16S sequencing (variable region V3-V4) and for shotgun analysis. All 16S samples were MiSeq sequenced as 2x250bp paired reads, the corresponding shotgun samples as 100bp paired reads. Shotgun analysis was performed for sample collected from 31/8/2016 until 5/10/2016 and only for EPI and 2.5x SECCCHI. The results showed that a peak of cyanobacteria was observed around 14.9/21.9 in the EPI (E) samples consistent with the high observed concentration of chlorophyll a. The lowest abundance of the cyanobacteria was in the SECCCHI (C) samples. Community composition of the overall cyanobacterial composition was also observed for proteobacteria and actinobacteria. Our result suggests that the major differences in bacterial community composition during the bloom are concentrated in the SECCCHI depth region while composition of the EPI zone is more or less constant. Cyanobacteria were found highly abundant in Lake Varese and are therefore likely responsible for the bloom. This hypothesis is also supported by the cyanotoxin data although complex and varying 18S metagenomes would be recommended in order to discard a possible contribution of phototrophic eukaryotes.

TU032
DNA metabarcoding demonstrates effects from copper at environmental concentrations on microbial diversity in marine periphyton biofilms
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Copper pollution is common in coastal areas. In particular, the use of copper-based antifouling paints on ships hulls elevates copper concentrations in these environments. This study assesses the effects of dissolved copper on community structure and function of marine periphyton biofilms. Microbial diversity and community function were assessed using 16S and 18S metabarcoding and 454 amplicon sequencing, targeting prokaryotic and eukaryotic organisms, respectively. Community functional studies were conducted as impacts on algal biomass, photosynthetic pigment profiles and primary production. Additionally, we studied Pollution-Induced Community Tolerance (PICT) using photosynthetic as the endpoint. Periphyton was exposed for 18 days to five copper concentrations, between 0.01 and 10 µM, in a semi-static test. The resulting 254 samples sequencing yielded 7.1 and 5.7 million high quality 16S and 18S reads, and the average numbers of 16S and 18S Operational Taxonomic Units among the samples were 9405 and 1242, respectively. Analysis of Unifrac distances showed that copper significantly changed the eukaryotic community structure at concentrations as low as 0.01 µM. The prokaryotic community structure was changed at slightly higher concentrations (0.06 µM). A total of 23 taxa, including species within the Proteobacteria_Bacteroidetes, Streptobacteria and Haeorobacteria classes, were identified as particularly sensitive to copper. Algal biomass, photosynthetic pigment profiles and primary production, were reduced at Cu concentrations of 0.06 µM and higher. PICT measurements confirmed that copper...
induced community tolerance in exposed communities. Taken together, these findings indicate that negative impacts from copper might be common in coastal ecosystems.

TU033
A Time-series Study of Soil Microbial Community Compositional and Functional Shift in Biodiesel vs. Petrodiesel Contaminated Soils

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Contamination of ecosystems by pesticides, pharmaceuticals and trace metals makes our approach a new useful bioindicators of ecotoxicity of contaminants in the region. Over the last three decades there has been a significant decline in marine pollution monitoring-related studies in South Africa. Very little research has been conducted to assess the prevalence of imposex in whelks and also very few studies have been conducted on comparisons between contaminants in different marine invertebrates at the same sites. The current study was conducted in July 2017 to compare metal bioconcentration between mussels (Mytilus galloprovincialis) and whelks (Busycon canaliculatum) as well as measure imposex prevalence in B. lanigera at Bloubergstrand, Granger Bay and Green Point, Cape Town, South Africa. This was done in order to identify suitable bioindicators of ecotoxicity by determining whether the mussels and whelks bioaccumulate metals in the same way and to assess imposex prevalence in whelks (as an indicator of tributyltin contamination). The bioconcentrations of metals (Al, Cu, Zn, Fe, Cr, Mn, Co, Cr, Ni, Mo, Cd and Pb) were measured in intertidal sediment, M. galloprovincialis and B. lanigera and imposex prevalence recorded in B. lanigera. Results showed that the highest prevalence of imposex in whelks and metal concentrations were recorded Granger Bay, an area of high boating activity. The most important result was that the whelks had higher bioconcentrations of metals than the mussels at all sites. Identifying biomonitor should be linked to purpose of investigation before selection of species, and mussels have been considered 'ideal' bioindicators of contamination in South Africa. Given the ubiquitous distribution of B. lanigera along the South African coast, which is not the case for M. galloprovincialis which only occurs on the west and south east of the country, the proposal is made that B. lanigera could be considered as alternative bioindicators of ecotoxicants of contaminants in the region.

Recent developments in environmental risk assessment for pollinators (P)

TU038
Behavioural effects of imidacloprid, a neonicotinoid insecticide, on bumblebees (bombus terrestris)

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Beekeepers are increasingly facing multiple and interacting threats. One of the threats that have received increased attention lately is neonicotinoids: a group of systemic neuro-active pesticides that disturb the transmission of signals in the insect’s nervous system. In just a few years neonicotinoids have become the most widely used insecticide in the world, and protect a variety of crops against invertebrate pest. Despite being used in relatively small quantities, several studies have shown sub-lethal effects of neonicotinoids on honeybees (Apis mellifera) and other bee species. Neonicotinoids are chronically deposited on crops and other plants, and are present in abiotic matrices such as pollen and honey. Studies have also shown that neonicotinoids can interfere with the foraging behaviour of honeybees. However, ecological and physiological traits vary among bee species and studies on honeybees may not provide satisfactory predictions for negative effects on other bee species. Using bumblebees, Bombus terrestris, the present study developed a new experimental method to quantify how chronic dietary exposure to the neonicotinoid imidacloprid affects learning, locomotor activity and consequently the ability to forage and thus pollinate in a non-Apis species. Bumblebees were exposed to three different dosages of imidacloprid through artificial nectar (sugar water), ranging from field realistic
levels (1 mg/L and 10 mg/L) to distinctly higher levels (100 mg/L) in a chronic exposure regime, lasting for eight days. To assess whether imidacloprid influences learning, the bumblebees' ability to discriminate between blue nectar-filled (rewarding) and yellow water-filled (non-rewarding) artificial flowers were tested systematically in a flying arena. The bumblebees were tracked by cameras, allowing for analysis of the flowers choices, locomotor activity and all the flowers visited during numerous, simultaneous foraging bouts. This study shows the successful application of a new method to track bumblebee behaviour. Further, the study shows that learning and locomotor activity are negatively affected, in a dose-dependent manner, when bumblebees are exposed to imidacloprid. Moreover, we show that field-realistic doses of imidacloprid have negative effects on bumblebees.

TU039 Sensitivity of honeybee larvae to PPs and impact analysis based on EFSA Bee GD
* R. Becker, BASF SE Agrarzentrum Limburgerhof; J. Lueckmann, Rifen GmbH
* on behalf of the ECPA NTA & Bee Working Group Based on EU Regulation 1107/2009/EC the current regulatory risk assessment on bees has to address the risk on honeybee larvae or honeybee brood. In July 2013 the European Food Safety Authority (EFSA) published a guidance document on the risk assessment of plant protection products on bees (EFSA 2013). This document is intended to provide guidance for notifiers and authorities in the context of the review of plant protection products (PPPs) and their active substances under Regulation (EC) 1107/2009 (EC 2009). The aim of this poster is to summarize available industry data, for active substances and formulated products on honey bee larvae testing, according to e.g. OECD 237 and OECD 239, in order to gain an overview of these results and the selectivity of different product groups. As a first step in the risk assessment, EFSA requires a screening step which consists of the calculation of risk quotients (RQs) for honey bee larvae. This considers exposure routes for the different PPPs, pesticides that could be used and off-field (PPPs) and as seed treatment (granules) scenarios. Where a substance or use should not pass one of the screening level risk quotients, EFSA offers the possibility for refinement in a tier I risk assessment. This includes the refinement of exposure estimates from the screening step and also additional exposure routes, such as the exposure to flowering weeds in field margins and adjacent flowering crops. The second objective of this poster is to evaluate the impact of the proposed screening and tier I risk assessments on the pass/fail rate of currently available active substances and formulated products which is an ability of the scheme to correctly identify compounds of potential concern and consequently screen out those of low concern. The aforementioned analysis follows the principles described in the ECPA impact analysis (Miles and Alix 2013) and compared the first approach with the outcome based on laboratory data. In

TU040 Honeybee brood studies according to Oomen and OECD GD 75: Is there a difference of the brood termination rate under semi-field and field conditions*
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* on behalf of the ICP-PR Bee Brood Working Group and the Bee Brood Working Group of the German AG Bienenschutz Based on EU Regulation 1107/2009/EC the current regulatory risk assessment on bees has to address the risk on honeybee brood. According to the guidance document on the risk assessment of plant protection products on bees (Apis mellifera, Bombus spp. and solitary bees) (EFSA 2014), both, the Oomen bee brood feeding test and the OECD Guidance Document 75 (OECD GD 75) under semi-field conditions rely on a realistic contact and oral exposure scenario to bees comprising contaminated nectar and pollen after overnighting of a bee attractive crop. As the evaluation of historical data from semi-field studies according to OECD GD 75 showed a strong variability of the control BTRs (Becker et al. 2015), the performance of OECD GD 75 bee brood studies under field conditions was regarded as an option to get more reliable BTR data (Becker et al. 2015, Giffard & Huart 2015). The present poster compares control BTRs from Oomen feeding studies with BTRs obtained from OECD 75 semi-field and field trials and considers explanations for observed variances. Moreover, the possibilities and limitations of the three methods will be discussed.

TU041 Does assessing of all brood cells of a hive reduce uncertainty and increase reliability of Semi-field honeybee brood studies (OECD GD 75)?
* I. Jäger, G. Gesior, M. Kleinhenz, B. Szczesniak, Eurofins Agroscience Services Ecotox GmbH; S. Knaebe, EAS Ecotox GmbH / Ecotox Field
The OECD guidance document 75 (2007) introduced a semi-field test method to assess the effects of PPPs on honeybee brood. The assessment of bee brood development over one brood cycle is conducted by mapping cells. It starts from the egg stage and the fate of individual cells is followed until hatch. For this purpose, pictures are taken at defined stages of the development cycle and compared to the development in separate control hives. Three parameters in regards to brood development are assessed and evaluated: brood termination rate (BTR) (number of the marked cells where a termination of the bee brood development was recorded, expressed as a percentage) and, as an indicator of the compensation of bee brood losses and brood index (an indicator of the bee brood development, facilitates a comparison between different treatments). Due to the high variability of BTRs within treatments and high control mortality in several studies no definite conclusions regarding effects on brood were possible (Pistorius et al. 2012). To address this variance, effort was taken by the ICP-PR Bee brood working group to develop a new approach, the semi-field tier I risk refinement of solitary bees. The goal of this tier I refinement is to use historical field and laboratory data to compare historical data and give recommendations for future testing (Pistorius et al. 2012, Becker et al. 2014). Despite fulfilling all recommendations, high variability in brood termination and high control mortality is still evident in some of the conducted studies. For those studies data interpretation and conclusions are questionable. According to Wang and Görlich (2017) one reason for the variability of brood termination rates is that the evaluation of brood development is based on a limited, defined number of cells containing eggs. According to their opinion results might be different if all cells would have been chosen. They suggested that it would reduce the uncertainty to a minimum. In this poster the recommendation of Wang and Görlich (2017) is used to find out if uncertainty can be reduced by evaluation of all cells. The brood data used were collected according to OECD 75 during a period of three years. The production of young queens, as the production of young queens is essential for the maintenance of a healthy bumble bee population. However, assessing the production of young queens in semi-field trials is challenging. Many variables influence the number of produced queens, such as the right timing for the termination of the study or the condition of the colonies at study start. Based on data collected in the past years, different strategies to reduce the variability in the production of young queens were evaluated. Also several parameters influencing this most important endpoint were analyzed. We tried to answer some open questions concerning the colony reproduction success, such as the experimental set-up can influence queen numbers and queen weights, how high the natural variation between colonies is and how the selection of bumble bee colonies for the studies can be improved.

TU043 Higher-tier risk refinement of solitary bee risks in the field - is the well-known ‘focal species’ concept a suitable approach?
* J. Lueckmann, M. Faupel, J. Ludwigs, Rifen GmbH
According to EFSA (2013) bumble bees and solitary bees have to be considered in addition to honey bees in the risk assessments. However, suitable testing methods in the lab are currently partly available only (e.g. for acute contact & oral bumble bee testing, acute contact solitary testing) or under development (e.g. chronic oral bumble bee testing, acute oral solitary testing). Regarding appropriate species for solitary bees EFSA (2013) proposes Osmia cornuta or O. bicolor as test organisms for the risk assessment, and higher-tier semi-field testing with Osmia as proposed by the ICP-PR Non-Api working group for these species. However, experiences from current field studies on Osmia show that exposure of adults and larvae is not necessarily given as these solitary bee species have a pronounced polylectic feeding behaviour that can result in a low exposure to a test substance (i.e. not being a real worst-case). In order to address this problem, the refinement of worst-case solitary bee risk assessments under realistic field conditions may be achieved by using a ‘focal species’ concept, where most appropriate focal solitary bee species can be identified to represent a worst-case choice per crop, application time and country/zone. Whereas this approach is well-known for bird and mammal risk assessment it has not been yet applied for

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solitary bees. Here, we present the idea of a ‘focal species’ concept for solitary bees, its needs, refinement options, advantages and limitations.

TU044

Non-Apis (Bombus terrestris) versus honeybee (Apis mellifera) acute oral and contact toxicity - Preliminary results of ECPA company data evaluation

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A preliminary data evaluation was conducted by ECPA companies to compare the sensitivity of bumblebees (Bombus terrestris) with the sensitivity of honeybees (Apis mellifera). For the evaluation about 70 data sets were available for contact exposure and about 50 data sets for oral exposure. The data sets comprised insecticides, fungicides, herbicides in about equal numbers plus a few other substances. The preliminary ECPA cutoff of LD50 valuing indicates lower or similar contact sensitivity of bumblebees vs. honeybees. Similarly, lower or similar oral sensitivity of bumblebees vs. honeybees was determined with one exception for an insecticide that indicated higher acute oral bumblebee sensitivity compared to honeybees. For this insecticide, higher tier data indicates no negative impact on bumblebees at the maximum intended use rate. Overall, a 10 ECPA company data evaluation indicates that bumblebees are not more sensitive than honeybees based on acute toxicity assessment.

TU045

Bumblebee (Bombus spp.) 10 day feeding laboratory test design: First results from an ICP-PR testing project

N. Exelr, Bayer AG, Crop Science Division; N. Hanewald, BASF SE / Ecotoxicology; C. Jenkins, Enivigo, H. Krueger, EAG Laboratories; A. Zicot, SynTech Research / Ecotoxicology; E. SOLER, TRIALCAMP SLU / Ecotoxicology; A. Molitor, Eurofins Agroscience Services GmbH; S. Vinall, Mambo-Tox Ltd; K. Amsel, BioChemagrar GmbH; S. Haupt, IBACON GmbH; S. Kimmel, Innovative Environmental Services (IES) Ltd / Ecotoxicology

A decline in some pollinator species has led to an increasing pressure on political decision makers and regulatory bodies and resulted in a changing of existing risk assessment paradigms and testing approaches. The published and already revised EFSA GD on the risk assessment of PPP on pollinators includes apart from the honeybees also bumblebees and solitary bees. In the need to address long term effects on bumblebees a Non-Apis Non-working group designed a ring test protocol to develop a first-tier chronic feeding test for bumblebees. Based on the recently published honeybee 10 day chronic feeding test guideline OECD 245 and the bumblebee acute oral toxicity test guideline OECD 247 a 10 day feeding test was set-up using dimethoate as reference substance. The response of adult Bombus spp. workers to the test chemical Dimethoate EC-400 (Perfektion) was evaluated within a 10 day feeding laboratory test design. The test item was provided ad libitum for a period of 10 days. During the exposure phase bumblebees are kept individually in cages – “single housing”. Bumblebees do not share food via trophallaxis and need to be fed individually. Furthermore, single housing prevents hierarchy fights (among the queen less BB workers) potentially introducing mortality. Mortality and behavioral abnormalities in the test groups were observed and recorded daily and compared to the untreated control groups. The endpoints calculated were: LC50 (median lethal concentration) and LDD50 (median lethal dietary dose) values after 10 days and if possible the NOEC (no observed effect concentration) and NOEDD (no observed effect dietary dose). First results indicate that with this method reproducible results were obtained. The mortality in the control groups seem not to exceed 15 % (evaluation currently ongoing) and the overall food consumption allowed for a proper evaluation of the intended endpoints.

TU046

Standardization of method to test toxicity on stingless bees

S. Knaeb, EAS Ecotox GmbH / Ecotox Field; N. Exeler, Bayer AG, Crop Science Division; L. Franke, J. Fricke, Eurofins Agroscience Services Ecotox GmbH / Ecotoxicology Field; M. Frommerberger, Julius Kuehn Institute; T. Jütte, Julius Kuehn Institute; S. Kimmel, Innovative Environmental Services (IES) Ltd / Ecotoxicology; O. Klein, Eurofins Agroscience Services Ecotox GmbH / Ecotox Field; J. Lueckmann, Rinfcon GmbH; H. Giffard, Testap; A. Rossbach, Tier3 Solutions GmbH / Field team; C.W. Schneider, BASF SE; A. Schnurr, BioChemagrar GmbH

The publication of the proposed EFSA risk assessment guidance document of plant protection products to honey bees. We test the Acute Contact Toxicity Test protocol of OECD guidelines (214) established to european honey bee indicate that a validated and workable methodology has been set up and a test guideline is within reach.

TU047

A method for a solitary bee (Osmia spp.) first tier acute oral laboratory test: an update

I. Roessink, Alterra / Environmental Risk Assessment; N. Hanewald, BASF SE / Ecotoxicology; C.W. Schneider, BASF SE; N. Exeler, Bayer AG, Crop Science Division; E. Noel, SynTech Research; A. Schnurr, BioChemagrar GmbH; A. Molitor, Eurofins Agroscience Services GmbH; E. SOLER, TRIALCAMP SLU / Ecotoxicology; S. Kimmel, Innovative Environmental Services (IES) Ltd. / Ecotoxicology; J. Van der Steen, Alveus AB Consultancy

The recently updated EFSA draft honey bee Guidance document also specifies other hymenopteran pollinators, like solitary bees and bumble bees, as groups to take into consideration when assessing the risk of plant protection products to pollinators. However no validated test protocol and consequently no extensive data set is available to compare sensitivities of other relevant pollinators to those of honey bees. Within the current project of the ICP-PR Non-Apis working group a start was made to develop a first-tier acute oral test for Osmia spp. bees. Based on the honey- and bumble bee guidelines OECD 213 and OECD 247 an acute oral test was designed using dimethoate as reference substance. Osmia bicornis and Osmia cornuta were housed individually and fed a known amount of test volume per dosage. First results indicate that with this method reproducible results were obtained. In these tests, control mortality never exceeded 12 percent. Furthermore, sensivities of O. bicornis and O. cornuta appeared to be rather similar, although O. cornuta showed a slightly less sensitive response, (which might be) due to its larger body weight. Hence, the L50-values after 96 hours ranging from 2.6 – 7.1 mg a.i./g bee indicate that a validated and workable methodology has been set up and a test guideline is within reach.

TU048

2 Years of Solitary Bee Semi-field Ring Testing and Final Conclusions (ICPPR Non-Apis Working Group)

S. Knaeb, EAS Ecotox GmbH / Ecotox Field; N. Exeler, Bayer AG, Crop Science Division; L. Franke, J. Fricke, Eurofins Agroscience Services Ecotox GmbH / Ecotoxicology Field; M. Frommerberger, Julius Kuehn Institut; T. Jütte, Julius Kuehn Institute; S. Kimmel, Innovative Environmental Services (IES) Ltd / Ecotoxicology; O. Klein, Eurofins Agroscience Services Ecotox GmbH / Ecotox Field; J. Lueckmann, Rinfcon GmbH; H. Giffard, Testap; A. Rossbach, Tier3 Solutions GmbH / Field team; C.W. Schneider, BASF SE; A. Schnurr, BioChemagrar GmbH

The publication of the proposed EFSA risk assessment guidance document of plant protection products for pollinators highlighted that there are no study designs for non-Apis pollinators available. Since no official guidelines exist for semi-field testing at present, a protocol was proposed and two years of ring tests were conducted in 2016 and 2017 to develop a general test set up. The ring test design was based on the ICP-PR Non-Apis Working group a semi-field was designed to develop a first-tier acute oral test for Osmia spp. bees. Based on the honey- and bumble bee guidelines OECD 213 and OECD 247 an acute oral test was designed using dimethoate as reference substance. Osmia bicornis and Osmia cornuta were housed individually and fed a known amount of test volume per dosage. First results indicate that with this method reproducible results were obtained. In these tests, control mortality never exceeded 12 percent. Furthermore, sensitivities of O. cornuta and O. bicornis appeared to be rather similar, although O. cornuta showed a slightly less sensitive response, (which might be) due to its larger body weight. Hence, the L50-values after 96 hours ranging from 2.6 – 7.1 mg a.i./g bee indicate that a validated and workable methodology has been set up and a test guideline is within reach.
termination rate during the larval development as well as the success of emergence of their progeny (F1-generations) in the following year. Based on the results of the ringtests over 2 years a draft protocol is available together with recommendations for the methodology needed. This includes how the cocoon incubation and hatching of bees can be synchronised with the onset of flowering, how fit solitary bees are out of season and which substance at what rate can be used as reference item for brood studies.

TT049 Predicting wild bee sensitivity to Acetylcholine Esterase (AChE) inhibitors utilizing a trait based phylogenetically controlled approach
T. Pamminger, BASF SE, Agrarzentrum Limburgerhof / Ecotoxicology; N. Hintermüller, BASF SE / Ecotoxicology; C.W. Schneider, BASF SE / J.C. Becker, New York State Dept. of Environmental Conservation / Biology; M. Bergtold, BASF SE

Plant protection products (PPP) play a vital role in modern agricultural practice. Nevertheless, their potential off-target effects on managed (e.g. Apsi mellifera) as well as wild (most non-Apis species) bees have emerged as an intensively discussed topic. In contrast, risk assessment to A. mellifera is often pure exposure scenarios in pollen to cover potential adverse effects of PPPs on non-Apis bee species. However, as robust and scientifically sound information regarding the sensitivity of non-Apis bee species are scarce the validity of this approach has been challenged. As a first step to address this question we have compiled a comparative data set of the Acetylcholine Esterase (AChE) inhibitors sensitivities of 21 bee species, covering five of seven currently recognized bee families. Method failures and data sets were complemented with information on bee bodyweight, a trait likely influencing bee sensitivity to PPP exposure. Our phylogenetic controlled analysis shows that bee bodyweight is a robust predictor of bee sensitivity to AChE inhibitors and confirms that A. mellifera is particular sensitive to this class of PPPs. In contrast, many stingless bee species, are comparatively resilient to AChE inhibitors, especially when controlling for body weight. We discuss the consequences of these findings in the context of the global non-Apis bee risk assessment debate in Europe and the Americas.

TT050 New approaches in testing of pollinator exposure under realistic conditions - Methods and recent experiences
M. Persianzehl, Tier3 Solutions GmbH; U. Zumkier, Tier3 Solutions GmbH / Regulatory Science; A. Rossbach, Tier3 Solutions GmbH / Field team; C. Wolf, Tier3 Solutions GmbH

With the growing concern for insect populations and an increased awareness of the importance of pollinators in the public opinion as well as in the regulatory context related to pesticides, we will show in the presented talk how the determination of residues as part of (semi-) field studies with bees in pollen, nectar and honey, studies on foraging behaviour as well as methods to adequately determine residues for non-standard uses, such as home and garden uses, ornamentals and granules with a slow release formula. The different methods are compared and advantages and potential pitfalls are illustrated.

TT052 Normative Instruction 02/2017 - Brazilian risk assessment of pesticides to bees
F. Viana-Silva, C. Dias, L. Borges, K. Cham, C. Tonelli, R. Oliveira, A. Alves, IBAMA / DIQUA CASGAQ; R. Rebelo, IBAMA / CCONP

An epidemiological study about an effect of neonicotinoids residues on honey bee colony survival in Japan
Y. Kameda, Chiba Institute of Technology / Creative Engineering; E. Fujita, K. Tachiya, Chiba Institute of Technology / Environment and Renewable Natural Resources (IBAMA) is responsible for the assessment of pesticides to bees in Brazil. The Environmenta...
for comparison with residue concentrations detected in pollen and nectar from treated and untreated crops. In 2016, a honey bee colony feeding study was conducted with thiamethoxam with the aim of providing a robust colony-level endpoint for comparison with residues in pollen and nectar. Analyses of the colony data indicate there were clear significant effects at the highest concentration of 100 µg/Kg for many colony parameters and overwintering survival. At 50 µg/Kg, despite a few treatment differences for pollen stores, overall colony strength and overwintering survival were not significantly different relative to the control, confirming the NOAEL as 50 µg/Kg. The NOEL was determined to be 37.5 µg/Kg. To assess the potential risk to honey bees from exposure to thiamethoxam and metabolite CGA322704 (clothianidin) residues in pollen and nectar, the NOEL and NOAEL can be compared to measured residues in treated or untreated crops. In a treated oilseed rape-exposure study (Pilling et al., 2013) the maximum thiamethoxam residues found in pollen and nectar were 1.0 µg/Kg and 3.0 µg/Kg, respectively. The residues of CGA322704 were below the 1.0 µg/Kg LOQ. In an on-going study, residues in pollen and nectar in untreated succeeding crops of sugar beet were also found to be low. The maximum thiamethoxam residues in pollen and nectar were 2.6 and 0.55 µg/Kg, respectively. A maximum CGA322704 residue of 6.3 µg/Kg was detected in pollen, while residues in nectar were less than the 1.0 µg/Kg LOQ. The colony NOEL and NOAEL concentrations are an order of magnitude greater than the maximum residues in succeeding crops and a treated crop. The colony NOEL and NOAEL provide the basis by which to evaluate the potential risk of thiamethoxam residues detected in pollen and nectar. It also provides additional support for the lack of effects reported in field studies following exposure of crops to levels of thiamethoxam in pollen and nectar of seed treated crops that are an order of magnitude lower than the no effect level observed in this study.

TU056 Alteration of the alternative splicing pattern in honeybees’ nervous system gene development as a tool to test pesticides toxicity


In recent years a number of population models have been developed for honeybees and have been used for pesticide risk assessment. While the in-hive development of honeybees is relatively well understood and can be validated relatively easily in models, the accurate estimation of exposure is more complex and more difficult to validate. In particular, foraging behaviour, which is included explicitly only in very few models, plays an integral role for exposure, since it determines to what extent foragers collect nectar or pollen from treated or untreated crops and other habitats, or if they find alternative food sources. Foraging behaviour is also tightly related to weather. We therefore evaluate how foraging behaviour can be implemented and validated in a honeybee model simulating natural conditions, with particular focus on the risk assessment of pesticides and on the protection goals formulated in the recently published honeybee guidance.

TU058 Modelling and validation of honeybee foraging behaviour for the pesticide risk assessment

M. Wang, WSC Scientific GmbH / Dept Efate, Modelling; C. Dietrich, WSC Scientific GmbH

In honeybee field studies EFSA recommended in its latest guidance that field studies should ensure that the 95th percentile exposure percentile is met. Practically, it is challenging to confirm where honeybees actually foraged. In recent years a variety of methods have been developed and tested to establish a kind of foraging maps, based on waggle dance observations analyses, harmonic radar or RFID chips. Most of these, however, can realistically be used only based on relatively few individual bees. We therefore explored options for an automated analysis of waggle dance in honeybees. The system should facilitate the use of standard hives and should be usable without a computer in the field. We evaluate the reliability of the method.

TU060 How to increase test power and understand risk in refined honeybee trials

A. Görlisch, WSC Scientific GmbH / Dept Efate, Modelling; C. Dietrich, WSC Scientific GmbH

For honeybee semi-field and field studies EFSA defined SPGs (specific protection goals) in its latest guidance document on the risk assessment of plant protection products (PPP) on bees. Detrimental effects on colony size as a result of PPPs should not exceed a 7% threshold to not endanger the fulfillment of the ecosystem services provided by honeybees. The measurement of effects on colony size as small as 7% is often difficult to achieve due to high uncertainty and variability both reducing the test power. By applying a modified field methodology and test design the test power can be increased substantially thus allowing to conduct field studies that are able to reach the SPGs. For the semi-field study colonies with sister queens of equal strength were used. From these a subset of colonies was selected based on hive assessments, which started approximately four weeks prior to exposure, by selecting those colonies that would be similar during the exposure phase. During the whole study the colony strength was assessed by photographing all bees in hives (all frames and walls). Additionally, to include also the number of foragers in the assessment, hives were weighted with and without bees. To avoid an influence of the time of the day on the number of foragers counted with photography all colonies were photographed in parallel at the same time of the day. All frames of all hives were also photographed to assess brood development and to obtain a full overview of the condition of each hive at each time point. It is shown that by applying a refined, new field methodology and test design for field studies on honeybees the
TT061 The potential for immune activation and possible consequences for bees upon exposure to microbial pest control agents

B. Jones, M. Whittaker, Applied Insect Science Ltd

Microbial pesticides are unlikely to cause disease in non-target insects due to a lack of specific pathogenicity. However, simply exposing the insect to a microbe has the potential to activate subclinical responses that can lead to colony level effects. For example, injection with a non-pathogenic, microbial immune elictor induces a massive antimicrobial peptide response in bumblebees and honey bees. This immune response lasts several days and is costly to maintain. These costs are demonstrable through trade-offs between immunity and other life-history traits such as learning and longevity. In addition, immune activation alters many aspects of normal colony functioning, such as changes in foraging activity, decreased queen attendance, modified feeding behaviour, increased production of sexuals and forced ejection. Crucially, many of these effects only become apparent in the colony, and show specificity between bumblebees and honey bees. The established paradigm uses laboratory trials as a ‘worst-case’ scenario before progression to higher tier field trials, which may mask the downstream immunological impacts on endpoints such as longevity. Evidence exists for immune activation in insects via oral exposure with non-pathogenic bacteria. We therefore propose the need to establish whether oral and cuticular contact with microbial pesticides can induce the immune system in bees. Should immune activation be confirmed in the laboratory in the absence of lethal effects, further field trials may be required to reveal the consequences within the colony.

Environmental effects of metals: Improvements to risk assessment by considering speciation and bioavailability (P)

TT062 Assessment of Levels of Some Heavy Metals in the Organs of West African Dwarf Goat and Beef Cattle in Ogbomoso, Nigeria

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The dangers inherent in the exposure to heavy metals present in food products especially meat, have aroused widespread concern for food safety and human health. With increasing human activities and anthropogenic pollution sources, there has been deposition of large amounts of various toxic metals in the food material which ultimately make their passage into the tissue. This study aims at assessing the levels of five heavy metals (lead, cadmium, zinc, copper and iron) in organs of West African dwarf goat and beef cattle slaughtered in Ogbomoso metropolis, Nigeria. Chevon and beef samples of heart, intestine, liver, muscle and tripe were collected from both sexes of two breeds of cattle and West African dwarf goats. Raw and cooked samples were digested and analysed using the method described by the Association of Official Analytical Chemistry. For each metal, higher concentrations were observed in the edible tissues compared to the reference and exposure fish, with the exception of As. For Cd and Cu, a major peak was observed at 27 minutes indicating that Se was not bound to MT but rather to a different protein. For Cd and Cu, a major peak was observed after a retention time of 16 minutes, corresponding to the retention of MT and 0.005 mg bioaccessible Bi/kg. Bismuth had little effect on phagocytic efficiency of adult earthworm coelomocytes. After 28 days, Bi concentrations in earthworm tissue increased up to 21.2 mg Bi/kg and reaching a stationary state at 212 mg Bi/kg of soil.

TT063 Assessment of subcellular metal-binding ligands in white suckers (Catostomus commersonii): are all the metals accumulated in the heat-stable fraction (HSP) detoxified by binding to metallothioneins

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Assessment of subcellular metal-binding ligands in white suckers (Catostomus commersonii) is important for environmental risk assessment and management. The study aimed to investigate the subcellular metal binding sites in the white sucker (Catostomus commersonii) utilizing the concept of stable proteins (HSP). The stable proteins were subjected to metallothionein fractionation, generally obtained after homogenization, differential centrifugation and heat-denaturation steps. It is normally hypothesized that metals present in the HSP fraction are detoxified. To confirm this hypothesis, the nature of the metal-binding ligands found in the HSP fraction needs to be determined. Thus, the aim of this work was to investigate the ligands binding metals (As, Cd, Cu and Sc) in the HSP fraction from hepatic cells of white suckers collected at a reference site, a heavily polluted site in a lake subject to multi-metallic contamination. After isolation of the HSP fraction, we used size exclusion chromatography coupled to an inductively coupled plasma mass spectrometer (SEC-ICP-MS) to separate biomolecules present in the HSP fraction and to quantify the associated metals. For each metal, higher concentrations were measured in the HSP fraction of the exposed fish than in the reference fish, but overall, metal-handling strategies did not vary between the reference and exposure fish, with the exception of As. For Cd and Cu, a major peak was observed after a retention time of 16 minutes, corresponding to the retention time of MT, suggesting that these two metals were reasonably well detoxified and regulated in these fish by binding to MT. In contrast, for Se, a major peak was observed at 27 min indicating that Se was not bound to MT but rather to a biomolecule with lower molecular weight. Finally, regarding As, two major peaks were observed in the reference fish (25 and 27.5 min), whereas in exposed fish a major peak was identified at 29.5 min, suggesting the potential induction of a specific ligand to bind As in exposed white suckers. For future work, the identification of the Se and As binding biomolecules would be of great interest to determine if these metals are detoxified or if, conversely, the biomolecules are metal-sensitive and their binding to Se or As represents a threat for the health of fish.

TT065 Assessment of Toxicological Impact of Anthropogenic activities on Onitsha Stretch of River Niger in Southeastern Nigeria

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The impact of anthropogenic activities on an urban stretch of a major river in Nigeria with respect to endocrine disrupting compounds and heavy metal concentrations was investigated. Three sampling points were selected along the Onitsha stretch of River Niger, based on the inputs of different tributaries into the river. Heavy metal contents of the water samples were analyzed after acid digestion, while the endocrine disrupting compounds were analyzed using liquid
chromatography. The result obtained showed that six heavy metals with varying concentrations were obtained in the order of Zn>Pb>Hg>Cd>Cr>Ni. The HPI and MI values were far above the critical values. Results also showed EDCs to include PAH, phthalates, PCDDs, PCDFs, PBDEs, bisphenol A and PCBs. This study established that Ontitsa stretch of River Niger contains varying concentrations of heavy metals and EDCs. The stretch of that river is highly polluted, and anthropogenic activities are highly impacting negatively on the river. The river therefore need to regulate the activities of people, especially the influx and disposal of pollutants into this surface water.

Tu066 Bioaccumulation, DNA damage and metallothionein expression in plants grown on heavy metal contaminated soil supplemented with sewage sludge

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Many anthropogenic activities have contributed to a release of contaminants, including heavy metals, into the environment. Since plants cannot leave polluted areas, it is, therefore, essential to possess a vast range of defence mechanisms that can reduce the toxic effects of heavy metals (HM). Contamination of soil and water with HM not only decreases the growth of plants but since metals can be accumulated in plant tissues they cause a severe threat to animals and humans the food chain. Identification of plants response mechanisms to contamination is becoming a prime objective in research since this knowledge can provide a solution for soil contamination and metal accumulation in plants. Studies on plants have demonstrated the ability of specific proteins - metallothioneins (MTs) to hyperaccumulate heavy metals, and play a significant role in their detoxification and overall oxidative stress. The physiological roles of MTs are not completely understood and much is still unknown concerning their characterization in many higher plants. This study was to evaluate the effects of fertilization of contaminated with HM soil by sewage sludge on the genotoxicity levels and the expression of metallothioneins in plants shoots and roots. The toxicity assessment was conducted using selected measurement endpoints: germination index, roots length, the severity of DNA damage, chromosome aberrations and the expression level of metallothioneins.

Sinapis alba L. was chosen as a model plant for this experiment. Plants were grown for 28 days in a growth chamber where they were exposed to soil contaminated by HM from metallurgical activities and to contaminated soil amended with different concentrations of sewage sludge. The study showed the effects of sewage sludge on the level of genotoxic effects caused by heavy metals as well as on MT expression. As such, a significant increase in the expression of MTs was observed in plants grown under metal stress. These differences showed statistically significant changes between related conditions which means that presented assay can be used as a sensitive stress marker for phytoremediation process.

Ti067 Chronic toxicity assessment of Ni contaminated rivers in Japan using Ceridophila dubia for development of biotic ligand model for Japanese surface waters

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Ni is one of industrial essential chemicals and have been widely detected in Japanese river. US and EU have already established the water quality standard/criteria for aquatic life protection; however, it is still under development in Japan. In metal toxicity assessment, bioavailability of metals is an important factor and Ni bioavailability models (i.e. biotic ligand model (BLM)) for both acute and chronic toxicity have already been available for plant, invertebrates, and fish. They were determined using the availability on the surface waters (hard water in general), which have different water chemistry from Japan (soft water, in general). Since water chemistry parameter (e.g. Ca, Mg, Na, K, pH, natural organic carbon) highly influence on metal toxicity, we should check applicability of the existing BLMs on Japanese surface waters or develop our original BLM based on the data of Japanese surface waters. To collect Ni toxicity data in surface waters, we collected 45 river water samples from Ni contaminated rivers all over Japan and conducted the daphnid reproduction test using Ceridophila dubia, which is one of the most sensitive species to Ni and recently came into use as test species to evaluate surface waters and industrial effluent in Japan. We used The Windermere Humic Aqueous Model (WHAM7) for speciation calculation. Ni toxicity were predicted using the existing chronic Ni bioavailability model for C. dubia established by De Schamphelaere et al. (2006). Except for uncontaminated upstream samples, the daphnids demonstrated typical toxic symptom of Ni (delayed lethal toxicity) and reproduction inhibition levels were correlated with Ni concentration suggesting that Ni is the representative toxicants in the collected samples. However, in several stations, other metals (such as Zn) may also contribute the toxicity thus we should carefully interpret the mixture toxicity.

Tu068 Comparing metallic elements in corals from South Africa and the Mascarene Basin

V. van der Schaff, North-West University / Unit for Environmental Sciences and Management; R. Choong Kwet Yive, University of Mauritius / Chemistry; H. Bouwman, North-West University / Unit for Environmental Science and Management

Coral reefs are one of the most bio-diverse biomes on earth. One of the many dangers that coral reefs face is the accumulation of metals and metalloids in skeletal and tissues of the colonies. No knowledge exists on the state of metal and metalloid contamination in corals from the Western Indian Ocean (WHO). Fragments of four soft- and five hard coral genera were collected from five sites in the WIO. Sodwana and Aliwal Shoal constituted the coastal sampling localities from South Africa. Three Mauritian outer-islands in the Mascarene basin (Agalega, Rodrigues, and St Brandon’s Atoll) were the selected oceanic sampling sites. Eighty-one coral fragments were collected and analysed for 31 metallic elements using ICP-MS. The corals collected from South Africa contained a higher concentration of most of the metals that were analysed compared with the Mascarene Island samples. Corals without symbiotic algae could only be collected from the South African reefs, and contained the highest concentration of metalloids. Soft corals exhibited a different relative composition pattern of metals than hard corals. Have been used to analyse as well. The use of different organisms in hard corals. Soft corals contained relatively higher concentrations of most of the post-transitional metals that were analysed. Sinularia is the coral genus with the most elements at the highest concentrations. Pocillopora from SBR had very high concentrations of Fe and Cr, possibly due to several shallow shipwrecks in the atoll. Most of the elements tested had lower concentrations in the WIO than in certain regions of the Great Barrier Reef and the Red Sea. Iron was consistently higher in all corals collected during this study than in corals from other studies. Some metals, such as Cu, Ni, and Cd, affect fertilization success of corals. Very high concentration of Ni was reported in Sinularia (1300 mg/kg dm) from Sodwana. As ocean temperature rises and ocean acidification increases, metals can become more bioavailable to corals, requiring further study.

Tu069 Cytochrome P450, fat and ageing: new insights into metal toxicology

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CYPs and fatty acid metabolism can unravel possible mechanisms of metal induced toxicity. Metallothioneins (MTs) to hyperaccumulate heavy metals, and play a significant role in their detoxification and overall oxidative stress. The physiological roles of MTs are not completely understood and much is still unknown concerning their characterization in many higher plants. This study was to evaluate the effects of metal contamination on the genotoxicity levels and the expression levels of metallothioneins in plants shoots and roots. The toxicity assessment was conducted using selected measurement endpoints: germination index, roots length, the severity of DNA damage, chromosome aberrations and the expression level of metallothioneins.

Sinapis alba L. was chosen as a model plant for this experiment. Plants were grown for 28 days in a growth chamber where they were exposed to soil contaminated by HM from metallurgical activities and to contaminated soil amended with different concentrations of sewage sludge. The study showed the effects of sewage sludge on the level of genotoxic effects caused by heavy metals as well as on MT expression. As such, a significant increase in the expression of MTs was observed in plants grown under metal stress. These differences showed statistically significant changes between related conditions which means that presented assay can be used as a sensitive stress marker for phytoremediation process.

Tu070 Determination of the effects of platinum in the oyster (Crassostrea gigas) using cell and tissue level biomarkers

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Purposes, metals entering aquatic ecosystems are regulated by water quality concentration in the environment, however human activity increases SQG values by the World Health Threshold element levels for the protection of aquatic life. Risk analysis using Agilent 7500c (Agilent Technologies, Tokyo, Japan) Inductively Coupled Plasma surface sediment from 15 sites in Lagos lagoon during the wet and dry season by an analysis. This study evaluates the level of contamination and potential ecological anomalies. Different histochemical parameters such as lipofuscin and neutral lipids were also measured and combined with metallography to detect the location and quantity of Pt. An increase of autometallurgical black silver deposits in T7 and T28 was detected for the highest exposure concentrations. 

On the other hand, neutral lipid levels showed a significant decrease at T28 for the exposure conditions Medium and High. Only minor and non-significant alterations occurred at histological level. This experiment has shown that short-term (28 days) exposure to relatively high Pt concentrations in seawater do not induce alterations at histological level. Neutral lipids were also measured and combined with autometallurgy. Different histochemical parameters such as lipofuscin and neutral lipids are impaired in oysters. Acknowledgements: Work funded by, Basque Government (ITB10-13), UPV/EHU (UFI 11/37), EU FP7 Ocean 2013.2 Project SChEMa (Project-Grant Agreement 614002), IdEx University of Bordeaux.

TU071 Ecological Risk Assessment of Trace Metal Contaminated Tropical Estuarine Sediment, Southwest Nigeria A. U. Sese, University of Lagos, Nigeria / Department of Marine Sciences; O.L. Chukwu, University of Lagos Nigeria / Marine Sciences; R. Naidu, The University of Newcastle upon Tyne / Global Centre for Environmental Remediation GCER, Faculty Science and Information Technology; M.M. Rahman, The University of Newcastle / Global Centre for Environmental Remediation GCER, Faculty of Science; S. Islam, The University of Newcastle / Global Centre for Environmental Remediation Faculty of Science and Information Technology.; E.O. Oyewo, Nigerian Institute of Oceanography and Marine Research / Victoria Island, Lagos Lagos state, N. Nigeria. The largest of the eight lagoons that make up the lagoon systems of Nigeria has been under intense pressure from several anthropogenic influences over the years. This study evaluates the level of contamination and potential ecological risk of trace metal (Cd, Co, Cr, Cu, Fe, Mn, Ni, Pb, and Zn) concentrations in surface sediment from 15 sites in Lagoons during the wet and dry season by an Agilent 7500c (Agilent Technologies, Tokyo, Japan) Inductively Coupled Plasma Mass Spectrometer. The Metals concentrations, with the exception of Cd and Pb, were below detection limits in the order Fe > Mn > Zn > Cu > Cr > Pb > Zn > Ni. Cd rarely exceeded threshold element levels for the protection of aquatic life. Risk analysis using contamination factors (CF) and Enrichment factor (EF) suggests very significant enrichment from Zn and Cd as well as a high degree of contamination (Cd) from Cd (16.88-21.56) at locations closest to urban runoff, industrial activity, domestic and solid waste dumps. Estimated pollution load index (PLI), geochemical accumulation (Igeo ? 0) and as well the applied sediment quality guidelines (SQG) values by the World Health Organization (WHO) and United States Environmental Protection Agency (USEPA) indicates low to moderate degree of contamination from sediment metals concentrations and the unlikely risks to ecological receptors during the study period.

TU072 Effects of culture medium on metal toxicity and new approach for ecotoxicology assessment G. Pasqual, Tohoku University / Civil and Environmental Engineering; I. Garcia, T. Torzecza, O. Nishimura, Tohoku University / Architecture Civil and Environmental Engineering. Pollution of aquatic ecosystems is a global problem affecting the biological continuity of living organisms. In this context, metals is a group of pollutants occurring naturally in the environment, however human activity increase its concentration in the environment. Therefore, for environment surveillance purposes, metals entering aquatic ecosysytems are regulated by water quality guidelines. The last one is based on the results obtained in toxicology tests using aquatic organisms, nonetheless the reported medium Effective Concentration (EC50) of a tested metal in a species widely varies. Here, our first purpose was to study the effects of culture medium on metal toxicity. Based on these results, our second purpose was to propose a new approach for the evaluation of metal toxicity on microalgae avoiding the interference of culture medium. In this study, we evaluated the toxicity of copper (Cu), lead (Pb) and zinc (Zn) on the microalgae Pseudokirchneriella subcapitata, since they are considered to be more sensitive to chemicals compared with other aquatic organisms such as fish. Cu and Zn were chosen for study metal species. A range of concentrations were play and observed for each metal to determine its toxicity. However, for the other study metal, Pb, any positive, biological function has not been reported. All tests were run in transparent microscope (96 wells), and pH of test solutions was adjusted at 6.5. The algae growth was determined measuring the fluorescence (433/685 nm). In the first experiment, the microalgae was exposed for 72 hours to each metal using three different types of culture medium, OECD modified OECD standard medium (mOECD) and Bold Basal Medium (BBM). In the second experiment, the microalgae was exposed in a simplified test medium (destilled water buffered with MOPS and NaOH) only for 6 hours, as nutrients available for algae were limited. In the first experiment condition, the EC50 after 72 hours were 140, 1200 and 289 μg/L for Cu, Pb and Zn in OECD medium, respectively, in mOECD, they were 34, 219 and 134 μg/L respectively and in BBM, they were >300 μg/L in all the cases. In the second experiment, the obtained EC50 after 6h were 150, 189 and 88 μg/L for Cu, Pb and Zn, respectively. The obtained EC50 of the metals differed between culture mediums. So the composition of culture medium affect the metal toxicity. Hence, we suggest that the simplified test medium may be an appropriate alternative to evaluate metal toxicity preventing interference of culture medium.

TU073 Environmental diagnosis of water and tilapia Oreochromis niloticus of the Tenango dam, Puebla, Mexico. M. Manoz-Najera, G. Barrera Escrigu, Universidad Autonoma Metropolitana Inlapalapa / Hidrobiología; P. Ramirez Romero, U.A.M. Inlapalapa / Hidrobiología Human population has seen the deterioration of resources derived from the overexploitation and contamination by anthropogenic activities, an example of this is water. Due to the growing demand for this resource, associated to population growth, industry, livestock, and agriculture, dams have been built to satisfy these needs. In Mexico, some of them were constructed in sites/areas that over time were declared protected natural areas. Population/in settlements on its banks to make use of the water, as well as of the organisms linked to these aquatic bodies, which represents economic sustenance for the inhabitants. On the other hand, it is common to use the water for various purposes, many of which contradict each other. Such is the case of the Tenango Dam, in Puebla, Mexico, which is used for fishing, irrigation, recreation and electric power generation, among other purposes. A study was carried to evaluate the quality of the dam's water and tilapia quality. Five field/trips were made in 2015. Physicochemical parameters were recorded: pH, dissolved oxygen and temperature; as well as nutrients: nitrates, nitrites, phosphorus; also metals: cadmium, chromium, copper and lead were determined in both water and tilapia. Results indicated that the physicochemical parameters are within Mexican admissible ranges. Nitrite and phosphorus exceeded the acceptable limit for urban use and protection of aquatic life. Lead and chromium in water exceeded the limits in four/collections, and tilapia, only in two of them. Cadmium and copper registered in water behaved similarly exceeding in two seasons the levels allowed by Mexican law, while in tilapia, cadmium only exceeded the acceptable limits for consumption in two seasons. Based on the concentrations of nutrients and metals, it is concluded that water from the Tenango dam is not suitable for urban use, nor for the protection of aquatic life and tilapia should not be consumed. These levels of contaminants could represent a risk to the life's associated with this artificial water body. The diverse uses and the absence of a management strategy have deteriorated the dam's water quality and also the tilapia as a resource associated with it; finally, this situation compromise the integrity of an aquatic body included in a site declared as a protected natural area.

TU074 Estimation of Target Hazard Quotients and Potential Health Risks of Some Heavy Metals from Lipsticks in Nigeria. O. Odebanji, Federal University of Technology / Department of Biotechnology; T. Oritoju, University of Nigeria Nsukka / human nutrition and dietetics Heavy metals have been implicated as a causal factor in literally any health problem including infertility and cancer. Their presence in most cosmetic products may pose more harm than envisaged. Lipsticks are common beautifying cosmetics used by young and matured ladies for the purpose of appearing attractive. However, the presence of heavy metals in most lipsticks may help to predict the possible risk associated with the use of these products. The main objective of this paper is to evaluate the hazard quotients of heavy metals due to daily ingestion or use of lipsticks among users and also to evaluate target cancer risks due to its use. This study was carried out in Wukari, Nigeria, samples of different lipsticks and lip glosses of many colours and texture were collected and analyzed for heavy metal contents (lead, Arsenic, Chromium, Cadmium and Mercury) using Atomic Absorption Spectrophotometer (AAS). The result of the mean concentration of the heavy metals are as follows; Lead, ranges between(2.65-7.40 ± 0.17) mg/kg;
Arsenic concentration range between 0.55-1.53 ± 0.26 mg/kg and chromium was 0.04-0.16 ±0.02) mg/kg. Cadmium and arsenic concentrations were below detectable limit of 0.001 mg/kg while mercury concentration ranges between 0.04-0.61 ± 0.01) mg/kg. Calculated target hazard quotient (THQ) was highest in mercury with the value of 0.56.99 and the lowest value was obtained in Arsenic with 1.43×10^{-3}. However, target cancer risk (TR) was highest for Lead with the value of 2.5×10^{-10}. The maximum value for cadmium was 1.53×10^{-3}. This shows that some lipsticks products popularly used in Nigeria contain high concentration of heavy metals such as Lead, Arsenic, Chromium, Cadmium and Mercury and they have high cancer risk. Therefore, public health awareness on the risk associated with the use of these cosmetic samples should be carried out.

TU075

Fatty acid profile of Cerastoderma edule and Scrobicularia plana affected by copper sulphate exposure
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At the past 30 years were recorded an intensive practice in the use of fertilizers and pesticides, mainly in the European Mediterranean region, that, in particular cases, exceeded the limits of regular legislations established by the European Union. The wide, rapid use of these chemicals is responsible for the pressure over agricultural fields near valuable ecologically coastal areas conducted to the implementation of monitoring plans to the recovering of aquatic ecosystems. Copper sulphate is used in industrial activities, but also it is much used in pesticides formulations, with application in agricultural activities, namely in rice farms to control pests. Studies reported that copper may affect biochemical processes, such lipid metabolism of some organisms and induced oxidative stress as a consequence of copper changes in fish. However, the effects of copper sulphate on the two marine bivalve species Cerastoderma edule and Scrobicularia plana when exposed to copper sulphate, considering small (medium body size = 1.97 cm and 3.47 cm, respectively) and big (medium body size = 2.45 cm and 4.20 cm, respectively) size classes. In a first phase experiments were exposed under laboratorial conditions to copper sulphate to determine lethal concentration; at a second phase, it was conducted to assess the effects of copper exposure on the nutritional quality of different size classes at the field and in the lab. Our results state C. edule is more sensitive to copper sulphate (LC50 = 0.818 (0.595–0.987) mg/L; 1.129 (0.968–1.289) mg/L, to big and small organisms, respectively) than S. plana (LC50 = 2.563 (2.229–2.903) mg/L; 4.705 (3.540–12.292) mg/L, to big and small organisms, respectively). Furthermore, the last one presents greater abundance and variety of FA and especially fatty acids (EPA), namely DHA and EPA, ratios than C. edule. Still, big size class of both bivalve species is the most affected by the contaminant.

TU076

Heavy metals in soil and vegetables of allotment gardens in the Cape Town, South Africa
M. Kweza, E. Wewers, T. Oosthuysen, T. Farrar, A. Giwa, Cape Peninsula University of Technology / Chemistry

Increased industrialization has resulted in an unprecedented dissemination of toxic substances, among which are heavy metals, in the environment. Heavy metals are persistent environmental contaminants which ultimately accumulates in soil with possible translocation into the tissue of vegetables, thereby posing a potential risk to human health. While most research focus on major agricultural areas, less attention is paid to the accumulation of heavy metals in home gardens, schools and rural areas where subsistence farming is increasingly used in South Africa as a means of poverty alleviation and increasing food security. This study was conducted to investigate the concentration of selected heavy metals in soil, water and vegetables. How do various test method conditions affect metal removal, using OECD method 29? What sediment characteristics affect metal removal and which are metals released into the water column under toxic conditions. The modifications include the addition of a small amount of sediment, and the inclusion of a resuspension event. We conducted a series of laboratory evaluations to address the following questions: Are copper (Cu) and nickel (Ni) removed from the water column of freshwater systems and if so, what is the rate of removal? How do various test method conditions affect metal removal, using OECD method 29? What sediment characteristics affect metal removal and which show a reasonable worst case (RWC) condition? What is the mechanism for metal removal, and are metals released into overlying waters upon subsequent resuspension? Method parameters evaluated included: sediment type and loading rate, pH control, metal loading rate, pre-incubation of sediment, and resuspension. Sediment loading rates included dissolved Cu, Ni, and Fe, dissolved oxygen (DO), pH and AVS-SEM of sediments. Multiple dried vs. non-dried sediments were tested in batch reactors for both 96 h and 28 d tests. Dry Buffalo River sediment, a sediment with reasonable worst-case properties for metal binding, typically removed 70% Cu and Ni from the water column at 1 mg/L loading. Incubated sediments removed metals significantly faster than non-incubated sediments (p < 0.03). Higher sediment loading rates removed metals faster as expected. Sediment type and loading rates affected pH, which started at 6.0. Cu removal (96 h) and resuspension (1 h post 96 h) resulted in no significant increase in Cu, but did elevate Fe concentrations. The results show that 70% of Ni and Cu is removed from the water.
column using this test modified OECD 29 test method, using a variety of sediments and conditions.

TU079 Modelling the chronic toxicity of copper to fish at low pH

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Bioavailability models account for the effects of water chemistry on metal toxicity to biota. They are a cornerstone of the environmental risk assessment of many trace metals, including copper. In this context, it has often been assumed that toxicity of dissolved copper to fish increases with decreasing pH. However, some studies show that this relationship may only be valid above pH 7. Below pH 7, the chronic toxicity of dissolved copper to fish seems to be independent of pH. Existing bioavailability models use the well-known mechanistic concept of the Biotic Ligand Model (BLM), but this modelling framework seems to have difficulties to reproduce the observed relationship of copper toxicity versus pH. This study was set up to refine the bioavailability models for chronic copper toxicity to fish, in order to better reflect the observed relationship between chronic copper toxicity and pH. The available chronic copper toxicity data to fish were reviewed. A new bioavailability model was developed using the concept of a generalized bioavailability model (gBAM). This semi-empirical model assumes a log-linear relationship between pH and effect concentrations (ECx) expressed as free cupric ion activity (Cu2+) and links it to the geochemical speciation model WHAM7 to predict toxicity on a dissolved copper basis. The existing bioavailability models were evaluated and compared to the newly developed gBAM. The underlying assumptions, advantages and limitations of each model are identified. Conclusions and implications for modelling copper bioavailability to fish are discussed.

TU080 Novel In-situ Toxicity Assessment of Sediment Capping Effectiveness in Deep Water

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A former mining site has been the subject of intensive restoration for the past few years, with significant focus on disconnecting mine spoils from groundwater and managing the quantity and quality of runoff. A remaining task is to ensure that concentrations of zinc in surface water of a large pit lake are reduced below water quality standards. An investigation was conducted to compare the efficacy of three methods of sediment capping for decreasing Zn dissolution during periods when the hypolimnion is anoxic and acidic (pH<5.5). Capping materials were selected based on results from laboratory batch testing and included AquaBlok, limestone, and limestone + bonechar. Experimental field tests implemented novel methodologies, using Limnocorals (LC) to isolate water columns above various capping treatments, simulating lake-mesocosms. Simultaneous in-situ and ex-situ toxicity testing were conducted using Daphnia magna, Hyalella azteca, and Chironomus dilutus. Test organisms were protected from temperature shock by pre-acclimatizing over 24 hrs and then deploying the test chambers in a Toxicity Assessment Container System (TACS), which protected the organisms from warm surface waters until reaching the bottom sediments and colder water. Test organisms were exposed to surficial sediments in reference LC or capping materials and overlying water. In-situ testing was conducted in waters and/or sediment cores collected from the bottom of each LC, and these tests were done at the same temperature as the in-situ TACS exposures (15 to 19°C, depending on deployment period). Results from in-situ testing demonstrated the usefulness of the TACS and provided similar results to the ex-situ testing. Preliminary results suggest organism survival is similar between in-situ capped and reference sediments; however, supplemental ex-situ analyses will help determine whether capping performance and toxicological response is indicative of site specific characteristics (sedimentation, sediment type, cap layer attenuation) and/or cap specific indices (permeability, adsorption, ion exchange capacity). Results provided for more effective decision-making, with reduced uncertainty, than standard laboratory and chemistry only approaches.

TU081 REEChEagE - Rare Earth Elements Ecotoxicology in a Changing Environment

H. Tien, Hamburg University of Applied Sciences/University of the West of England

H. Tien, Hamburg University of Applied Sciences / Life Sciences

REEChEagE focuses on the anthropogenic release of the rare earth elements (REE) lanthanum and gadolinium to the environment and their potential risk within ecosystems. REE are increasingly applied e.g. in green technology, and consequently also emitted to the environment. But there is a diversity of potential polluting sources of which little is known, and no regulatory environmental framework for inclusions exists so far. In addition, a review by Herrmann et al. (2016) demonstrated the considerable lack of reliable data for La toxicity in the aquatic environment. Considering their future use, release, and environmental fate, an evaluation of environmental risk from lanthanum and gadolinium will have to be based on information on exposure pathways, exposure and effect concentrations. The project REEChEagE addresses these topics in the following ways: (1) by studying the toxicity to aquatic organisms. Results on ecotoxic responses obtained for Alivibrio fisheri and Rhapidoceolus subcapitata so far are in the same range as literature data, and show a higher toxicity of Gd compared to La. Effect concentrations are of the same magnitudes as for cadmium. (2) by substance flow analysis (SFA) for La and Gd, exemplarily performed for Germany. Information has been collected from published work for a variety of potential sources for La and Gd in rivers and lakes. Additionally, water and sediment samples have been analysed at specific locations. Current data point to wastewater and specialized industries as prominent sources of emission. (3) by investigating the impact of changing environmental conditions (pH, redox, salinity) on the bioavailability of particle bound La and Gd. In a microcosm, a battery of miniaturized biotests were applied to monitor the toxicity responses in overlying water and sediment. This includes tests with Alivibrio fisheri, Vibrio proteolyticus Arthrobacter gelbrini and especially Daphnia magna. Additionally, speciation and bioavailability of the La and Gd are examined following a procedure by Simpson et al. (2014), applying a cascade of different filters and a chelating resin. The presented poster will depict the results of the microcosm experiments along with the information on bioavailability based on biotests and speciation data.

TU088 Sediment characteristics of natural and anthropogenic origin and their parabolic association with benthic macroinvertebrates in a minimally affected river in South Africa.

C. Wolmarans, H. Pienaar, G. van Niekerk, North-West University School of Biological Sciences / Zoology

Sediment characteristics generally entail metals, minerals, organic content, elements, particle size and composition and pH. The origin of metals in sediments may originate from anthropogenic activities including mining, industries, agriculture as well as aerial deposition. Limited attention is however given to the contribution that natural occurring phenomena play in the concentration of metals in the sediment. Weathering of minerals originating from the primary lithology can on the one hand add to the metals in sediments and on the other hand to the particle size composition. Although it is well known that chemical pollutants, due to anthropogenic impacts, act as a major determinant for the macroinvertebrates composition in surface waters the influence of the above mentioned components in a pristine river is less known. The aim of this investigation was firstly to determine the sediment characteristics and secondly to establish which of these characteristics have a significant impact on the macroinvertebrate community structure in the Mantobeni river. A South Africa collection consisting of 4 replicates of substrate from various sites, dried and sieved using an Endocott dry-sieving system to collect fractions <2000µm and <50µm. The total sediment samples >2000µm and clay fraction samples, 50µm were subjected to metal, scanning electron microscopy and minerals by X-ray diffraction analyses. Element anlyses were done by means of an FEI Quantas 230 ESEM ESEM microscope equipped with an integrated Oxford Inca X-Max 20 EDS. Macroinvertebrates present in the benthos were collected for 15 minutes using a standard sweep net, preserved in 90% ethanol and identified up to family level. RDA redundancy analysis was constructed to investigate the distribution of macroinvertebrates Forty two families of which the vast majority associated with particles >2000µm, were found. Sediment particle sizes <2000µm had a detrimental effect on the biodiversity. No significant correlation was demonstrated in the variation in electrical conductivity and both diversity and abundance of macroinvertebrates. Although relatively high concentrations of selected metals were present in the sediment, it was largely from geological origin and most probably not bioavailable. Therefore, it can be concluded that, under these conditions, sediment particle size, played the decisive role on the distribution and abundance of macroinvertebrate taxa.

TU083 The effect of copper sulphate on the antioxidants enzymes activity of two size classes of Cerastoderma edule

A. D. Mesquita, Department of Biology & CESAM - University of Aveiro / Department of Biology; S. M. Maciel, University of Coimbra; E. C. Marques, University of Coimbra / MARE, Dep. of Life Sciences, Coimbra University; F. Gonçalves, University of Aveiro / Department of Biology and CESAM; A. M. Gonçalves, MARE, Dep. of Life Sciences, Coimbra University/Biologia Department & CESAM, Aveiro University

Anthropogenic activities, such as agriculture or industrial activities are the main sources of heavy metal pollution contributing to the degradation of the aquatic ecosystems, and thus affecting the living organisms of the aquatic systems. Copper is often released into the aquatic systems, and may affect these ecosystems and their communities. Copper sulphate is a copper-based formulation, used in agriculture practices to control pests. The main aim of this study is to determine the effects of copper in the antioxidant defence system of an important commercial bivalve species, Cerastoderma edule in two size classes. In this work was observed the behaviour activity of the organisms during the exposure time to copper sulphate and subsequently it was determined the antioxidant enzymatic activities of GST, GRed and GPx in the muscle tissue (foot). Moreover, lipid peroxidation was evaluated
through thiobarbituric acid reactive substances (TBARS) measurement in the muscle tissue. The results showed changes in the behaviour and enzymatic activity at the different copper sulphate concentrations to both size classes. Moreover, according to TBARS levels, lipid peroxidation possibly occurred on the big size class of C. edule. The muscle tissue (foot) showed to be a good tissue to use in biochemical analysis to detect response to the exposure to toxics.

TU084 The impact of single metals and mixtures in nature: a microcosm experiment M. Van Ginneken, University of Antwerp, Dept. Biology / Biology (SPHERE); R. Blust, L. Bervoets, University of Antwerp / Department of Biology (SPHERE Research Group)

Environmental and ecological risks of metal-contaminated systems remains an important challenge. While laboratory experiments with metal mixture exposure are receiving more attention in the literature, little research has examined the interaction of natural stressors with metal mixtures. In the lab, we already performed experiments on *Axelius aquaticus*, exposing this freshwater isopod to a combination of metal mixtures and temperature stress. This way we could study effects on the individual level and relate metal accumulation to relevant sublethal endpoints (e.g., growth rate, feeding rate). The present study, a microcosm experiment in a greenhouse, was designed to gain more insight into the effects of these metals on populations and communities. Small ecosystems with several species of macroinvertebrates were exposed to Cd, Cu, and Pb and a mixture of these three metals under semi-natural conditions. In each bucket, we placed *Axelius aquaticus*, *Daphnia magna*, *Cryptocoryne reniformis*, with different pH-values. Soil samples with wide range of properties were spiked with Pb(NO₃)₂ at 9 concentrations of Pb to determine the effects of soil properties on Pb bioavailability and toxicity to *E. crypticus*. Survival and reproduction after 21 days exposure was related to total, 0.01 M CaCl₂ extractable and porewater Pb concentrations in the soil and internal Pb concentrations in the surviving animals. pHeq, pHeq, and pHporewater decreased with increasing total Pb concentration for the soils, but pH decrease was much stronger for the soils with lower CEC and OC contents. Sorption of Pb from the CaCl₂ extracts could be well described by a Freundlich isotherm (R² = 0.96-0.99) and Freundlich sorption constant Kₛ increased linearly with increasing cation exchange capacity (CEC) (R² = 0.86) or organic carbon content (OC) (R² = 0.76). Pb bioaccumulation in the enchytraeids was soil-dependent, but differences between soils almost disappeared when relating Pb bioaccumulation to available Pb concentration in soils. Toxicity values varied greatly among soils, with median lethal concentrations (LC₅₀) based on total Pb concentrations ranging from 246 to >3092 mg Pb/kg dry soil. LC₅₀ on the basis of total Pb concentration increased linearly with increasing CEC (R² = 0.70-0.90) or pHporewater (R² = 0.87-0.94). The differences in Pb toxicity among soils could be explained from CaCl₂ extractable Pb concentrations in the soils (R² = 0.97) and internal Pb concentrations (R² = 0.97). Median effective concentrations (EC₅₀) based on total Pb concentrations varied 12-fold among soils from 81 to 1008 mg Pb/kg dry soil. EC₅₀ on the basis of total Pb concentrations increased linearly with increasing pHporewater (R² = 0.70-0.94). The variation in EC₅₀ was best explained by differences in the CaCl₂ extractable Pb concentrations in the soils (R² = 0.94). In general, pH was an important soil property affecting LC₅₀, EC₅₀ and internal Pb concentrations in enchytraeids, as Pb availability, internal Pb, mortality and reproduction were inversely related to soil pH. Soil properties should be taken into account during the ecologic risk assessment of metals in contaminated soils.

TU085 The influence of soil properties on lead bioavailability and toxicity to *Enchytraeus crypticus* L. Zhang, VU University Amsterdam / Animal Ecology; C. van Gestel, Vrije Universiteit Amsterdam / Ecological Science

Soil properties are important factors modifying metal bioavailability to soil organisms and subsequently affecting the metal toxicity. The present study aimed at investigating the bioavailability and toxicity of lead to the potworm *Enchytraeus crypticus*. Soils with wide range of properties were spiked with Pb(NO₃)₂ at 9 concentrations of Pb to determine the effects of soil properties on Pb bioavailability and toxicity to *E. crypticus*. Survival and reproduction after 21 days exposure was related to total, 0.01 M CaCl₂ extractable and porewater Pb concentrations in the soil and internal Pb concentrations in the surviving animals. pHeq, pHeq, and pHporewater decreased with increasing total Pb concentration for the soils, but pH decrease was much stronger for the soils with lower CEC and OC contents. Sorption of Pb from the CaCl₂ extracts could be well described by a Freundlich isotherm (R² = 0.96-0.99) and Freundlich sorption constant Kₛ increased linearly with increasing cation exchange capacity (CEC) (R² = 0.86) or organic carbon content (OC) (R² = 0.76). Pb bioaccumulation in the enchytraeids was soil-dependent, but differences between soils almost disappeared when relating Pb bioaccumulation to available Pb concentration in soils. Toxicity values varied greatly among soils, with median lethal concentrations (LC₅₀) based on total Pb concentrations ranging from 246 to >3092 mg Pb/kg dry soil. LC₅₀ on the basis of total Pb concentration increased linearly with increasing CEC (R² = 0.70-0.90) or pHporewater (R² = 0.87-0.94). The differences in Pb toxicity among soils could be explained from CaCl₂ extractable Pb concentrations in the soils (R² = 0.97) and internal Pb concentrations (R² = 0.97). Median effective concentrations (EC₅₀) based on total Pb concentrations varied 12-fold among soils from 81 to 1008 mg Pb/kg dry soil. EC₅₀ on the basis of total Pb concentrations increased linearly with increasing pHporewater (R² = 0.70-0.94). The variation in EC₅₀ was best explained by differences in the CaCl₂ extractable Pb concentrations in the soils (R² = 0.94). In general, pH was an important soil property affecting LC₅₀, EC₅₀ and internal Pb concentrations in enchytraeids, as Pb availability, internal Pb, mortality and reproduction were inversely related to soil pH. Soil properties should be taken into account during the ecologic risk assessment of metals in contaminated soils.

TU086 Toxicity evaluation of soils sampled in the vicinity of an aluminium smelter in Montenegro using the Ames, Biomiminescence and DR-LUC biosayss A. Perovic, University of Montenegro, Faculty of Natural-sciences and Mathematics / Biology; S. Perovic, J. Vukic, University of Montenegro Faculty of Naturalsciences and Mathematics; D. Sukovic, Centar for Ecotoxicological Investigations; H.A. Leslie, Institute for Environmental Studies VU Amsterdam

This study conducted as a part of the national project ECOTOXI that main topic was testing applicability of several bioassays in assessment of cause-effect relation between levels of organic environmental pollutants in soils and its toxic and mutagenic response on samples organic extracts. Samples was collected in vicinity of Aluminum Plant Podgorica and pools of red sludge in Zeta plain. This area with intensive industrial activity is also reach with agriculture and is just in 5km distance from Podgorica (Capital of Montenegro). Waters of several rivers in this area, which are significantly influenced by municipal wastewaters of tree cities, are abundantly used for watering crops in area. We tested mutagenic potential of samples organic extracts in the Ames test, on bacterial strain *Salmonella typhimurium* TA98, acute toxicity on bioluminescence of bacteria *Vibrio fisheri* and concentrations of possible dioxins present in the samples by DR-Luc test on rat H4IE hepatoma cell line. The obtained results indicate a strong mutagenic effect of organic pollutants mixture in tree samples collected near the Aluminum Plant and pools of red sludge, what was significantly in correlation with the recorded concentrations of dioxins in the DR-Luc test and with measured concentrations of polycyclic aromatic hydrocarbons, which exceeded the maximum allowable prescribed concentrations. Two of tree samples, with high response, were in agricultural area. Even if it is a clear trend of decrease of mutagenic effects as well as reduction of the concentration of dioxins and PAHs with increasing distance of sampling sites from the Aluminum Plant and pools of red sludge, almost all samples showed a certain elevated level of mutagenic activity, which may be a consequence of the impact of multiple sources.

Safe by Design: responsible and innovative research for safe and sustainable chemistry (P)

TU087 In silico approaches to screen and design safer chemicals E. Papa, A. Sangion, P. Gramatica, University of Insubria / Department of Theoretical and Applied Sciences (DiSTA)

The prohibitive economic and social cost of testing, necessary to provide extensive information on fate and effects of existing chemicals to humans and the environment, highlights the need to focus on rational and safe design of chemicals before synthesis (i.e. Safe by Design – SbD approach). This approach applies the principle of green chemistry “Design safer chemicals and products, and is useful to prevent hazardous substances from being developed and entering the environment, as well as to build safer alternatives to existing hazardous chemicals. While in the last decades computational chemistry and in silico models have been widely and successfully applied in the design of drugs with desirable pharmacological activity, these strategies have not yet been applied extensively in the design of sustainable, “safe by design” industrial chemicals as well as no real guidelines exist at the regulatory level. Modelling approaches based on Quantitative Structure-Activity Relationships (QSARs) rely on the assumption that biological activities/properties of chemicals are intrinsically dependent on the molecular structure. Endpoints like for instance toxicities, physico-chemical properties as well as biotic and abiotic degradations can be predicted starting from models based on molecular descriptors of the chemical structure, which serve as basis to develop the SbD approach. Therefore, in silico strategies such as the aforementioned QSAR (and QSAR-like) models and multivariate analysis (MVA) can be successfully applied to screen untested properties of large sets of chemicals in order to identify potentially hazardous compounds or safer alternatives. In this poster we show different examples of QSAR models mainly implemented in the software Q SAR and available in the freely distributed QSARINS-Chem module to screen “safe” from “unsafe” compounds on the basis of different endpoints of scientific and regulatory interest. Different classes of emerging pollutants were investigated using in silico models, such as Flame Retardants (FR), Personal Care Products and Pharmaceuticals (PPCPs) and nanoparticles. All the presented strategies support the identification of safer alternatives to chemicals that are screened by QSAR as undesirable from their molecular structure. The QSAR approach, applied for SbD before the chemical synthesis, provides concrete opportunities to increase the sustainable use of chemicals and to reduce the need for a posteriori remedial actions.

TU088 Application of chemometric methods and QSAR models to support pesticide risk assessment starting form ecotoxicological datasets E. Galimberti, ICPS International Centre for Pesticides and Health Risk Assessment, Milan; F. Tagliarini, Public Health University of Trieste, Italy; A. Moretto, Università degli Studi di Milano; E. Papa, University of Insubria / Department of Theoretical and Applied Sciences (DiSTA)

Recently the International Center for Pesticides and Health Risk Assessment (ICPS) is involved in the European safety Authority (EFSA) the aim of the project was to investigate the comparability of the EC, approach (Effect Concentration affecting a percentage x of test organisms) to the current NOEC approach (No Observed Effect
Concentration), both derived from chronic and long-term studies of a data sets of 70 active substances of plant protection products (PPP). The new Regulation for the authoriztion of PPPs requires that ecotoxicological endpoint values, derived from chronic or long-term studies submitted by the Applicant, are reported as EC\textsubscript{10} or EC\textsubscript{20} as well as NOEC. NOEC endpoints have been recently criticized since their values strongly depends on the experimental study design, whereas EC\textsubscript{20} values are considered more appropriate and they take into account concentration-response curve. Ecotoxicological data gathered from 70 active substances’ approval dossiers were collected and stored into a database, and then analyzed to derive NOEC. Adequate statistical models were selected and used to calculate EC\textsubscript{50}, E\textsubscript{C50}, and EC\textsubscript{20} with confidence intervals. In the present work, quantitative methods and models based on Structure-Activity Relationships (i.e. QSARs) were used to validate the Effect Concentrations of the active substances of the selected pesticides, and to predict missing data. The whole approach is mainly oriented to the aquatic environment, and can provide useful information to screen the potential undesired toxic effects of new pesticides, and of alternatives to existing active substances, starting just from the chemical structure.

TU089

Influence of coatings in the bioaccumulation of TiO\textsubscript{2} and CeO\textsubscript{2} nanoparticles in rainbow trout

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In the framework of FP7 Project GUIDEnano we investigated the effect of different metal nanoparticle (NP) coatings (sintetized from PlasmaChem GmbH, Germany) on the the toxicity and bioaccumulation of these NPs trying to establish some relationships between coatings and the related effects. Bioaccumulation studies with rainbow trout have been performed for CeO\textsubscript{2} NPs and TiO\textsubscript{2} NPs of 4-8 nm uncoated and coated with citrate or polyethylene glycol phosphoric acid ester (PEG). OECD Test Guideline (TG) 305 (diet administration) has been followed. Fish (5±1 g weight) were fed for 10 days with a diet spiked with 100 mg/kg of the NPs dispersed in water. A control group fed with pellets containing the vehicle (water) was tested in parallel. This uptake phase was followed by a depuration phase of 42 days. Whole fish, stomach and intestine were collected at different time points (0, 10, 11, 17, 24, 38 and 52 days). In addition at the end of the uptake and depuration phase liver and gills were collected. The levels of the metals in these tissues were measured by inductively coupled plasma mass spectrometry after an acid digestion. During the treatment and depuration phase, no signs of toxicity and no differences in fish growth or in the hepatosomatic index among groups were recorded. At the end of the uptake phase levels of Ti could be measured in stomach, gills and liver without differences among TiO\textsubscript{2} NPs. A difference was observed for the uncoated NP for which Ti levels in the fish were higher than for the other coated NPs. Ti levels reached basal values already in the first day of depuration indicating a very fast elimination of these NPs from the organism. Higher levels of Ce with respect to the control group could be measured at the end of the uptake phase in stomach, intestine and gills but not in liver. Ce levels were found in fish treated with the coated NPs but not in the group treated with the uncoated NPs. Levels of Ce could be measured the first day of depuration in stomach and intestine of fish treated with CeO\textsubscript{2} NPs coated with citrate whereas Ce was only detected in the stomach of fish treated with CeO\textsubscript{2} NPs uncoated or coated with PEG. After 7 days of depuration, Ce residues reached basal levels indicating a lack of accumulation of these CeO\textsubscript{2} NPs. These results indicate a different behavior for the CeO\textsubscript{2} NPs and TiO\textsubscript{2} NPs. No relationship could be observed between the coating and the observed effects.

Acknowledgements: EU FP7 project 604387 GUIDEnano.

TU090

Collodial characterization of nano-enabled products for the restoration of works of art: environmental fate of nano-ingredients

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The development of highly innovative techniques and technologies for artworks preservation is providing conservators with new engineered nanomaterials (ENM) and ENM-based formulations that can enhance performance and technical sustainability of art materials [1]. However, the human health and environmental impacts that may potentially emerge from these new materials and/or techniques are still little known and requires an adequate assessment and management of potential risks [2,3]. ENM formulations are increasingly preferred for conservation interventions compared to the corresponding bulk materials formulations because of their small size and enormous specific surface area that favour their interaction with the material to be conserved/restored. But the small size, coupled with their capacity to adsorb biologically active compounds, can contribute to the transport to reach sub-cellular locations leading to potentially higher localized concentrations and toxicity. A lot of factors such as size, shape, surface coating and the fact that these particles are subject to fast alteration, make complicated the elucidation of the interaction mechanisms of these nanomaterials with the artefact material and the surrounding environment including the nano-bio interaction. In this study, in the frame of the EU FP7 NanoNoTox project, innovative nano-enabled formulations for the conservation and restoration of modern and contemporary artworks have been provided, following a Safe-by-Design (SbD) approach. The safety of the new formulations was investigated by applying both EU CLP self-classification approach for mixtures (ECHA, 2017) and experimental in vivo and in vitro ecotoxicological tests. In order to better understand the key interactions occurring between ENMs and the biological medium of the tests, the colloidal characterization of the new formulations was performed by means of Dynamic Light Scattering (DLS) and Centrifugal Separation Analysis (CSA) techniques. Moreover, possible releases from outdoor conserved works of art were also investigated by immersion tests, simulating the exposure of the treated materials to the worst applicable environmental conditions (e.g. rain, humidity, temperature).

TU091

Considerations for Safe Innovation: The Case of Graphene

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In the context of, in the frame of the EU FP7 project 604387 GUIDEnano we investigated the effect of different metal nanoparticle (NP) coatings (sintetized from PlasmaChem GmbH, Germany) on the the toxicity and bioaccumulation of these NPs trying to establish some relationships between coatings and the related effects. Bioaccumulation studies with rainbow trout have been performed for CeO\textsubscript{2} NPs and TiO\textsubscript{2} NPs of 4-8 nm uncoated and coated with citrate or polyethylene glycol phosphoric acid ester (PEG). OECD Test Guideline (TG) 305 (diet administration) has been followed. Fish (5±1 g weight) were fed for 10 days with a diet spiked with 100 mg/kg of the NPs dispersed in water. A control group fed with pellets containing the vehicle (water) was tested in parallel. This uptake phase was followed by a depuration phase of 42 days. Whole fish, stomach and intestine were collected at different time points (0, 10, 11, 17, 24, 38 and 52 days). In addition at the end of the uptake and depuration phase liver and gills were collected. The levels of the metals in these tissues were measured by inductively coupled plasma mass spectrometry after an acid digestion. During the treatment and depuration phase, no signs of toxicity and no differences in fish growth or in the hepatosomatic index among groups were recorded. At the end of the uptake phase levels of Ti could be measured in stomach, gills and liver without differences among TiO\textsubscript{2} NPs. A difference was observed for the uncoated NP for which Ti levels in the fish were higher than for the other coated NPs. Ti levels reached basal values already in the first day of depuration indicating a very fast elimination of these NPs from the organism. Higher levels of Ce with respect to the control group could be measured at the end of the uptake phase in stomach, intestine and gills but not in liver. Ce levels were found in fish treated with the coated NPs but not in the group treated with the uncoated NPs. Levels of Ce could be measured the first day of depuration in stomach and intestine of fish treated with CeO\textsubscript{2} NPs coated with citrate whereas Ce was only detected in the stomach of fish treated with CeO\textsubscript{2} NPs uncoated or coated with PEG. After 7 days of depuration, Ce residues reached basal levels indicating a lack of accumulation of these CeO\textsubscript{2} NPs. These results indicate a different behavior for the CeO\textsubscript{2} NPs and TiO\textsubscript{2} NPs. No relationship could be observed between the coating and the observed effects.

Acknowledgements: EU FP7 project 604387 GUIDEnano.

TU092

Safer-by-Design framework for supporting small and medium enterprises early in sustainable innovation for nanomedicine

M. Schnitz, C. Soni, EMMA Technology & Society Lab

One of the hot topics in nanomedicine is the use of nanobiomaterials for drug delivery. On the one hand, nanobiomaterials have various expected advantages compared to its bulk material: 1) decreased doses, 2) possibility to cross biological barriers, 3) increased drug efficacy, 4) reduction of side effects, and 5) targeted drug delivery. On the other hand, the nanosize brings new challenges for risk assessment of nanomedicine: i) it is a complex, and combines knowledge from different fields. It is at the junction among pharma, medtech, biotech, nanotech and chemical companies which are important economic and social player in Europe. In this context, the GoNanoBioMat project aims to facilitate SMEs in Europe in the decision making for developing and producing safer and sustainable polymeric nanobiomaterials for drug delivery. To do so, the consortium designed a ‘Safe-by-Design’ framework that aims to support the innovations occurring in this field and to find out which are the critical quality attributes (link between physico-chemical properties and toxicity, product safety, quality and purity). Furthermore, there are difficulties in reproducing environmental and human health experiments for assessing the related
risks and having batch-to-batch uniformity. Finally, notifying bodies are behind development because of the uncertainties arising from this field. Therefore, it seemed important to include in the framework the following aspects: safe material's design, human health and environmental risks, manufacturing, storage and transport and the regulations related to the topic at hand. At the end of the project, the Safer-by-Design framework will be used as a structural backbone for creating nano-specific guidelines for medicine. These guidelines aim to facilitate the communication among the different stakeholders in the value chain and with regulators for safe and sustainable innovation.

TU093

Review of the applicability of early-stage sustainability methods integrating toxicology and environmental assessment

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The chemical industry strives for the development of bio-based alternatives for present fossil-based chemicals driven by the transition to a bio-based economy. Key in this transition is "safe and sustainable by design", which means safety and sustainability are taken into account at the earliest possible development stages. Many sustainability assessment methods are developed for this purpose. The aim of this study is to evaluate a selection of 12 early-stage methods (ESMs), their applicability, the relevance for bio-based chemicals and the coherence of their outcomes, using bio-based lactic acid as a retrospective case study. The selected methods contain at least one of the following themes: energy, climate change, eutrophication, land use, human toxicity and ecotoxicity. These six themes were considered as most relevant for the case of bio-based lactic acid. The selected early-stage methods were compared to the right hotspots concerning energy and climate change, which is promising for application during process design. In general, the selected ESMs define simple environmental and toxicity indicators that have lower data requirements and are faster to implement than full assessment methods. However, the results they provide have intrinsically a higher level of uncertainty. Besides, the ESMs existing in the literature do not meet important criteria for utility. They are often not clear in the definitions of the environmental and toxicity indicators neither transparent in background data sources and not up-to-date. Important limitations of the selected ESMs are 1) narrow life cycle scopes (excluding the environmental impacts of material and biomass feedstock production) and 2) omission of some environmental aspects relevant to bio-based materials throughout their life cycle in general. Within this study, we pinpoint limitations and positive aspects of several early-stage sustainability methods. Based on this exercise we identify and propose successful elements of existing methods to be included in a framework that supports the assessment of safety and sustainability in early development phase.

TU094

Liquid organic hydrogen carriers (LOHC) - comparative hazard assessment

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Renewable energy stored in LOHC systems could replace fossil fuels yet their environmental impacts are largely unknown. This technology is still relatively new, and early development phase. The formulation of biofuels for obtaining information on potential hazardous properties. These bioanalytical tools can assess adverse effects of many substances on various organisms and endpoints and thus provide a rapid and reliable screening of potential biofuels for identification of potentially harmful biofuel candidates at a very early stage of product development. The testing strategy is part of a framework proposed by the new discipline of "Green Toxicology" which strives to move safety considerations of newly developed chemicals to the earliest possible moment of its lifecycle. Aquatic toxicity is considered as one important ecological endpoint relevant for biofuels. Therefore, the investigation of aquatic toxicity of promising biofuel candidates focused on acute immobilisation of Daphnia magna and acute embryotoxicity and teratogenicity of Danio rerio. Moreover, genotoxicity of the biofuel candidates was also investigated in the Micronucleus assay with V-79 cells to assess the potential effects on human health. This study focuses on the investigation of two biofuel derived fuel candidates: 1-Octanol and 1-Butanone. Both substances are considered very promising alternative fuels. The toxicity testing revealed a very low acute and developmental toxicity for 2-Butanone compared to 1-Octanol. 2-Butanone induced acute toxicity and genotoxicity in concentrations >2 g/L and even teratogenic effects were found at 822 mg/L. 1-Octanol did induce effects in concentrations between 7-15 mg/L. The overall results indicate that 2-Butanone is not harmful for aquatic organisms and should be focused in the further biofuel development. For a further integration of this screening approach in the biofuel development, more biofuel candidates can be investigated and, thus, more detailed information on their potential toxicity can support the development and production of green biofuels. This work was performed as part of the Research Cluster "Refuelling the fuels from biomass" funded by the Excellence Initiative by the German federal and state governments to promote science and research at German universities.

TU095

Investigation of the toxic effects of new mixtures of deuterateic solvents (DES) on the environment and human health

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The development of environmentally benign and green synthetic protocols, due to the high concern over climate change, has been central to the need to find greener, readily biodegradable and low cost solvents. This new concept of green chemistry has recently led to the synthesis of Ionic Liquids (ILs), from which have evolved in few years the deuterated solvents (DES).[1] These compounds are obtained mixing two components: a quaternary ammonium salt (e.g. ChCl) with different hydrogen bond donors, in such a ratio that the resulting substance has a higher hydrophobicity as it is the case for diesel oil. Octanol and 2-Butanone compared to 1-Octanol. Butanone induced acute toxicity and genotoxicity in concentrations >2 g/L and even teratogenic effects were found at 822 mg/L. 1-Octanol did induce effects in concentrations between 7-15 mg/L. The overall results indicate that 2-Butanone is not harmful for aquatic organisms and should be focused in the further biofuel development. For a further integration of this screening approach in the biofuel development, more biofuel candidates can be investigated and, thus, more detailed information on their potential toxicity can support the development and production of green biofuels. This work was performed as part of the Research Cluster "Refuelling the fuels from biomass" funded by the Excellence Initiative by the German federal and state governments to promote science and research at German universities.

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New frontiers in Life Cycle Inventory data collection and modelling (P)

TI/097 Predicting environmentally beneficial production pathways for chemicals with thermal networks
J. Kleinendijk, RWTH Aachen University, Institute of Technical Thermodynamics / Institute of Technical Thermodynamics; M.R. Tillmanns, M. Sternberg, RWTH Aachen University / Institute of Technical Thermodynamics; A. Bardow, RWTH Aachen University

Life Cycle Assessment (LCA) has gained wide acceptance as methodology to evaluate the environmental impact of chemical processes. However, LCA requires detailed data on mass and energy balances. Data is usually limited for processes in stages of early development. For these cases, predictive LCA approaches are required. Current predictive LCA approaches employ solely molecular descriptors to estimate the environmental impacts of products. Thus, the choice between different production pathways towards the same molecule cannot be resolved.

Therefore, we propose a neural network-based approach that uses both molecular and process descriptors. The resulting neural network is able to distinguish between various production pathways for the same product while still employing only data available at early stages of development such as stoichiometry. We estimate 5 impact categories including, e.g., cumulative energy demand (CED) or climate change (CC). The novel approach is compared to a neural network trained with molecular descriptors only. The results show that integrating process descriptors increases the coefficient of determination from 0.37 to 0.65 and from 0.39 to 0.65 for CED and CC, respectively. The route-specific prediction is illustrated for methanol production from CO₂ versus natural gas. It is shown that neural network models can serve as an initial screening tool for identifying environmentally beneficial new production pathways.

TI/098 A Study on the development of Food LCI DB and PCR for estimating environmental footprint in South Korea
S. Cho, SMAnt-ECO / Sustainable Business Div.; S. An, S. Seol, I. Kim, SMAnt Eco Corporation

As a result of the Paris Climate Convention adopted in December 2015, 195 countries in the world were required to implement greenhouse gas reduction, and Korea also proposed a 37% reduction target compared to BAU. In accordance with the environmental regulations of developed countries, Korea is also not free from environmental regulations at the time of preparation for active response. Under the Single Market of the EU, product environmental regulations starting from automobiles in 2000 expanded to include food in 2020, requiring the disclosure of high-quality environmental information on foods. As a result, the development of a high-quality environmental information database is accelerated, and the EU has created an ILCD Data Network to induce DB registration in each country. However, Korea’s agricultural and livestock LCA DB does not meet the environmental regulations in each country. Therefore, the development of a database that enables data on major domestic food exports to Europe and to use the common protocol and food - specific guidelines (PCR) to estimate environmental footprint, and aims to obtain EPD certification of food. To do this, we benchmarked the protocol and PCR for the Korean conditions by examining the cases of the calculation guidelines of the developed countries. In the future, it is meaningful to construct a database that can be used as basic data for obtaining PEF certification for foods exported to Europe and overseas.

TI/099 Transition from ILCD To Environmental Footprint: changes in the database structure, format, nomenclature, methods and other adaptations.
S. Fazio, EC-JRC; O. Diaconu, JRC European Commission; R. Pant, European Commission / Institute for Environment and Sustainability; S. Sala, European Commission - Joint Research Centre / Bioeconomy unit

In 2013 a Communication from the Commission to the European Parliament (COM/2013/0196) established the Environmental Footprint (EF) scheme. The communication methodology, nomenclature and recommended Life Cycle Impact Assessment (LCIA) Methods have been partly changed and adapted to fulfill the scope. Beyond that, the reference data contained in the ILCD package was found to contain some format, syntax and conceptual errors which were inherited from several data providers over time. Therefore a new database has been developed. Errors have been fixed, new files have been developed, and redundant or obsolete files have been deleted. The content of this presentation represents a synthesis, recalling general considerations or decisions, that have been applied for specific impact categories, and technical details with respect to each impact category, documenting specific choices made when implementing the characterization factors as well as tools and methods that have been implemented or will be implemented in the near future. Furthermore, a list of changes made from the ILCD to the EF package, beyond the LCIA methods recommended (i.e. Elementary Flows, Flow Properties, Unit Groups) and complementary objects defining the compliance (i.e. stylesheets, schemas) are described, in a change log file available through the EFLCA website. Among the above mentioned at once the overall changes occurred in the ILCD-EF transition phase can be resumed as following: - 1242 obsolete or wrong elementary flows have been deleted / mapped - 560 new elementary flows have been created - Around 55.000 characterisation factors are different (this is mainly due to the introduction of new methods, and regionalization of some of them) - 37 duplicated flows have been eliminated - 275 wrongly categorized flows have been assigned to the proper category - 218 wrong or useless flow properties have been deleted and mapped to the remaining ones - 35 unit groups have been deleted, one new has been created.

TI/100 New tools for Environmental Footprint data checking and sharing: Soda4LCA, ILCD validator and Registry for the node management
S. Fazio, EC-JRC; O. Diaconu, JRC European Commission; O. Kusche, OkworX; R. Pant, European Commission / Institute for Environment and Sustainability; S. Sala, European Commission - Joint Research Centre / Bioeconomy unit

Several tools for Life Cycle Inventory data development, validation, sharing and registration to the Life Cycle Data Network (LCDN) have been released by the EC since 2013 and improved after the official launch of the LCDN (2014). All those tools were originally meant for the International Life Cycle Data (ILCD) scheme. Since 2013 after a specific EC Communication (COM(2013)/0196) the Environmental Footprint (EF) scheme has been developed and led to significant changes in the structure of the ILCD/LCDN. During the development of EF compliant data, the tools had to be adapted and improved to fulfill new requirements. Particularly the following tools and software have been changed:
- ILCD validation tool: software for the compliance assessment of datasets (format syntax, archive structure, nomenclature, links and orphaned items, categorization, etc.) - sodalca software for distributing data based on the ILCD data format, with search and management functions, including the data registration in the LCDN - LCDN registry: online registration facility that can be accessed individually and merged, if the same data from different nodes running on soda4lca, and meant to make available only fully compliant data (while the nodes can host also intermediate data) - The changes that have been applied can be summarized as follows: - ILCD validation tool: additional validation profiles added for EF scheme. Checks against different parameters for Elementary Flows, location IDs, new LCIA methods, Flow Properties, Unit Groups and schemas, according to the changes made in the DB structure - soda4LCA: new access profiles are available for data stocks. The developer can now select entire data stocks and restrict the access only to authorized users. The entire data stock can be now downloaded directly, while before it was possible only at the single dataset level. The registration form includes a statement for the use of data within the EF framework. Declaration of compliance in the registration phase and possibility of multiple registration in more than one registry at once (a dataset can be both ILCD and EF compliant and therefore registered in two registries with different compliances declared). - LCDN registry: a dedicated registry has been developed for EF, with new functions. The compliance scheme is now visible (before was implicit since only ILCD was possible), possibility to register entire batches of data at once (before each single dataset had to be registered manually). Search interface improved.

TI/101 Improving the consistency and the accuracy of water inventories of chemical sites in PlasticEurope LCIs in the perspective of the applicability of the improved assessment methods
M. Baier, thinkstep / LCA; G. Castelan, PlasticsEurope / LCA

The ISO 14046 standard has clarified the definitions of water use and of water consumption which is the part of water withdrawn from a drainage basin not returned back to the same drainage basin. These only definitions have enabled PlasticsEurope to fix a number of important inconsistencies in its eco-profile protocols, where use and consumption were sometimes used interchangeably. In this short term action, in perspective of enabling the application of the latest consensus water assessment method AWARE to the water consumption, PlasticsEurope and thinkstep have collaborated to improve the consistency of the data collection phase of water flows for the various projects of the program. For the purpose of good water management in a chemical plant, plant managers need to have a good knowledge of all the water flows inputs, their origin (lake, river, public supply underground...), their treatment, in what equipment or process they are employed like for example in a cooling towers or being injected in the chemical process like in steam cracking. They must know the post-use-treatment and where all the outputs
end (back to the river, evaporated, in the public sewage network, in the product…). For the purpose of consistency of the Life Cycle inventory phase, it is then very important to report these collected operational flows in the ILCD input and output flows the right and same way whoever the LCA practitioner is. That location matters when it comes to quality of water footprint in order to go LCA. The presentation elaborates on the various operational water footprint in a chemical plant and the link to the life cycle inventory phase of ILCD flow name. The result PlasticsEurope methodology for calculating eco-profiles. It is expandable or adaptable to all kind of industrial sites. A similar work would need to be conducted on all datasets so that the consistency of water inventory gets improved in both foreground and background data, enabling a better comparability of water footprint in order to go LCA. The presentation aims to add LCA water exports and scientists as well as people applying the water methods in practice to exchange on challenges, relevancy of aspects and to align on a continuous improvement of water data, regionalization efforts and method improvement in the future, to inspire broad application.

TU102 Methodological improvements by dynamic approaches for the life cycle assessments of buildings K. Negishi, CSTB; L. Barna, INS A Toulouse / LISPB; Y. Pigné, Université de Havre; T. Navarrete-Gutierrez, LIST; N. Schioppu, A. Lebert, CSTB; T. Gibon, Luxembourg Institute of Science and Technology (LIST)/Environmental Research and Innovation (ERI); B. Boro, Luxembourg Institute of Science and Technology (LIST)/Environmental Research and Innovation (ERI).

Nowadays in France, environmental and energy rules for the construction sector are based on environmental performances of products assessed through LCA methodology. However, the actual practice of LCA is lacking of temporal dimension whereas the temporal evolution during the long lifetime span of buildings has non negligible impact on overall LCA results. A new framework of LCA method was recently proposed by L.Barna et al (2016) and A.Shimako et al (2017) with a particular attention to the process and supply chains (web tool DyPLCA, http://dyplca.pigne.org/), aiming at calculating time dependent environmental interventions and the related impacts of toxicity and climate change. The aim of this study is to investigate the environmental performances over a large time span of two low-energy single houses, one on concrete and one on timber. The time dimension was integrated on both LCA steps (LCI and LCIA) using the framework cited above. The implementation of dynamic LCA took several steps. Buildings life cycles were first modeled in SimaPro 8.02 with ecoinvent 2.2 to calculate the conventional LCI. The calculated technological and environmental interventions matrix was then used with DyPLCA web tool for temporal LCI calculation. The temporal characterization of the product system considered two parts. Temporal characteristics of the foreground system were related to the building construction, materials replacement and renovation activities during the building’s life time of 100 years. The temporal characteristics of background processes were previously integrated in a dedicated database and used with DyPLCA tool. The temporal LCI is then a result of environmental interventions distributed in time, then was used for climate change impact calculation in function of time. Two indicators were calculated in function of time: mean temperature change and radiative forcing. The new method allows considering fossil and biogenic carbon for climate change proposes without clamping on to fixed characterization factors and time horizon and without compensation as done in conventional method. The use of dynamic LCI methodology provides a more consistent and complete understanding and a more consistent analysis of environmental impacts of buildings. However, simulation time and memory usage for dynamic LCI calculation can be a principal limitation for the practice of dynamic LCA.

TU103 Carbon footprint from Brazilian soybeans based on spatially-explicit life cycle inventories, including land use change N. Escobar, University of Bonn / Institute for Food and Resource Economics IRL; J. Godar, Stockholm Environmental Institute.

That location matters when it comes to quantifying environmental impacts of agricultural commodities based on the increasing importance of the LCA literature. Authors tackle the influence of spatial variability by capturing differences in agricultural practices, transport options and industrial processing sites in the life cycle inventory (LCI). This information is, however, incomplete when quantifying impacts of agricultural commodities that are produced in large amounts and traded worldwide, e.g. soybean. Despite the efforts from the Input Output methodology to compare these impacts, the data usually requires the use of aggregated resource consumption and emission data for environmental extensions, which provides little detail on the technological and locative factors contributing to overall ecological footprints. From the LCA perspective, total nation’s supply is made of thousands of individual life cycles from farm to gate, for which LCI data is not frequently available. The Trace platform allows for real-world pathways of several internationally traded commodities to be annually mapped, from producing regions to destination countries. In view of its great potential for LCA purposes, our goal is to implement a carbon footprint module able to deliver results on C02eq. emissions associated, on the one hand, to annual production of soybean supplied from Brazil for the period 2010-2015; on the other hand, to every individual supply chain embodied in the whole supply of seed, oil, and cake to the international market. These include the following life cycle stages: land use change (LUC), soybean farming, domestic transport, export, and crushing, dealing with allocation challenges. In this way, our approach represents the convergence between top-down Multi-Regional IO analysis and bottom-up Attributional LCA. Preliminary results highlight the relevance of the approach that adds environmental information to the entirety of the supply chain, mainly LUC, for which considering sub-national scales is crucial in the quantification of climate change impacts. Outcomes support the argument that importing countries of soybean-based commodities should take responsibility on deforestation and associated carbon emissions, provided that spatial explicit data is available. This transparency tool is meant to provide science-based evidence to the ongoing debate on global responsibility, while assisting supply chain management and governance decisions.

TU104 Carbon Footprint Projections for Japan Using Computable General Equilibrium models Y. Ichisugi, Tokyo City University; T. Masui, National Institute for Environmental Studies; N. Isubo, Tokyo City University

In 2015, Science Based Targets (SBT) has been paid attention to the world. The targets adopted by companies reducing greenhouse gas (GHG) emissions to keep global temperature below 2 degrees increase from that of preindustrial revolution. Approximately 300 companies in the world declared to follow their targets. The target to the mitigation would be based on the calculated results of Integrated Assessment Models (IAM) such as Asia Pacific Integrated Model (AIM), Integrated Model to Assess the Global Environmental (IMAGE). However, these results usually don’t consider the entire supply chain, because of the differences of the aims of application. In contrast, Life Cycle Assessment (LCA) considers the entire life cycle chain. On the other hand, DyPLCA tool is a similar work would need to be conducted on all datasets so that the consistency of water inventory gets improved in both foreground and background data, enabling a better comparability of water footprint in order to go LCA. The presentation aims to add LCA water exports and scientists as well as people applying the water methods in practice to exchange on challenges, relevancy of aspects and to align on a continuous improvement of water data, regionalization efforts and method improvement in the future, to inspire broad application.
The added value of using invertebrate species in ecotoxicology: new insights for environmental risk assessment (P)

E. Vernon, The University of Plymouth / School of Biological & Marine Sciences; J.T. Smith, University of Portsmouth / School of Earth and Environmental Sciences; A.N. Jha, Plymouth University / Biological Sciences

1. Introduction

The aquatic environment is the primary recipient of anthropogenic pollution, including radionuclides. Despite the growing concern over presence of radionuclides in the aquatic environment, there has been only limited studies to determine potential biological responses in aquatic invertebrates following exposure to environmentally realistic levels of radiation doses. This integrated study determines and compares tissue specific bioaccumulation, doses delivered and induced biological damage in two species of mussels, the freshwater species Dreissena polymorpha (DP) and marine Mytilus galloprovincialis (MG), following exposures to an important radionuclide, phosphorus-32 (\(^{32}\)P).

2. Materials and methods

The study involved 10 day exposures of mussels to \(10^{12}\) P of varying dose rates (i.e. 0.10, 1.0 and 10.0 mGy/d) taking into account a current no-effect screening value of 0.24 mGy/d (European Commission). The first set of studies determined \(^{32}\)P accumulation in specific mussel tissues (i.e. adductor muscle, digestive gland, mantle, gills and ‘other’), internal mussel water (water inside the mantle cavity), shell and faecal matter using scintillation techniques. From this bioaccumulation study, we were able to highlight key tissues of interest; the digestive gland for example, received the greatest proportion of \(^{32}\)P independent of mussel species. In the next set of studies, a suite of biological responses on mussel gonads were investigated in digestive gland and gill cells. This included the induction of DNA damage (Comet assay) and repair response (Gamma-HAX), the induction of micronuclei (MN) and the expression of key stress related genes (i.e. SOD, CAT, GST, HSP70/90).

3. Results and discussion

Our findings highlighted DNA damage and MN induction at radiation doses as low as in 0.1 mGy/day in digestive gland (MN = also in gill) in both species, below the screening benchmark. Furthermore, compared to fresh water (FP), marine bivalve (MG) displayed increased internal DNA damage (both tissues) across all \(^{32}\)P treatments. This study highlights that (a) radionuclide activity concentrations in a biological system cannot be predicted by surrounding environmental media (b) with regards to bioaccumulation of waterborne contaminants, whole body measurements may mask the tissue specific nature of radionuclide uptake, and (c) the importance of adopting a multi species, multi biomarker approach when assessing the possible effect of contaminants in the aquatic environment.

TU109

Endocrine disruption in Mytilus galloprovincialis: Is ethinylestradiol a vitellogenin inducer?

L. Fernández González, P. Sanchez Marín, University of Vigo / Ecology and Animal Biology; G. Grueiro Noche, S. Muniategui Lorenzo, University of A Coruña / Analytical Chemistry Department; A. P Diz, University of Vigo / Biochemistry Genetics and Immunology; r. beiras, University of Vigo / Toralla marine sciences station (ecimar), Vitizegocénos (Vtg), the egg-yolk precursor in female oviparous animals, is a common biomarker of estrogenicity widely used as an indicator of endocrine disruption in aquatic environments. Nevertheless, in the case of mussels, it is still unclear if the synthesis of Vtg is regulated by steroid hormones as in the case of vertebrates. In the case of the synthetic hormone ethinylestradiol (EE2) the results of the studies are inconclusive. The aim of this work is to verify whether the estrogen in Mytilus galloprovincialis can induce the production of Vtg in Mytilus galloprovincialis at two exposure times and different nutritional regimes. For that, we used a shotgun label-free proteomics approach by high resolution LC-MS/MS to identify and quantify Vtg in mussels gonads. In this way, we can verify if the energetic balance is a key confusing factor in Vtg production. The relationship between the maturity state of mussels and their Vtg levels was studied as another possible confusing factor. Mussels from uncleaned area in Galicia (Spain) were collected in autumn/winter, corresponding with early gametogenesis stage. Mussels were exposed during 4 and 24 days to 100 ng L\(^{-1}\) of EE2 to assess whether Vtg synthesis was induced by EE2. During exposure, mussels were fed three times per week with two different regimes: a low regime (equivalent to 0.29 % of mussel dry weight per day), or with a high regime (equivalent to 5.55 % of mussel dry weight per day), representing negative and positive energy balance respectively. For the low feeding regime, shotgun proteomics identified an detected Vtg only in female gonads. The results showed an increase in Vtg levels in mussels exposed for 4 days to 100 ng L\(^{-1}\) of EE2 compared to the solvent control, although this increase was not statistically significant. In mussels exposed for 24 days to 100 ng L\(^{-1}\), Vtg levels were significantly higher than in mussels exposed to a solvent control. These results suggest that EE2 does not induce Vtg in M. galloprovincialis. However, it is possible that Vtg synthesis was impaired by the fact that organisms were in negative energy balance. The results of the experiments done at a higher feeding regime (currently being analysed) will confirm this result. A significant correlation was found between Vtg levels and the maturation state of female mussels, indicating that maturation state is a confounding factor for the application of Vtg levels in endocrine disruption studies.
Integrating natural processes in environmental hazard assessments of the oil sands
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The Athabasca oil sands deposits in northern Alberta, Canada are a naturally occurring mixture of bitumen, sand, clay and other minerals. Bitumen, which is a heavy and extremely viscous oil, is mined and then subsequently refined to produce gasoline, diesel and other hydrocarbon-based products. Moreover, the naturally occurring Athabasca Oil sands deposits are a source of both physical and chemical stressors to regional rivers that flow through the deposit. Physical stress on aquatic biota from natural bitumen results from hillslope erosion processes and slumping of material into the rivers, while chemical stress arises from bitumen-derived contaminants entering the waters. To fully understand the ecological and cumulative effects of oil sands mining activities on aquatic ecosystem water quality and associated biological structure and function, there is a need to evaluate the effects of naturally occurring bitumen in the aquatic environment. The main objective of this study was to evaluate the possible ecotoxicological effects associated with the slumping of river bank material (i.e. oil sands deposit that naturally enters the river systems through fluvial geomorphological processes). A series of inter-related laboratory ecotoxicological assays were conducted using both acute and delayed effects on the offspring and the population dynamics from four different sources in regional rivers (SP, ATB, STB and ELLs). All ecotoxicological results were complemented with the chemical analysis of metals, naphthenic acids (NAs) and polycyclic aromatic hydrocarbons (PAHs) to understand the possible effects that this material will induce when in contact with aquatic systems. All tested organisms responded negatively to the presence of oil sands deposit in the water through their toxic effect to DNA methylation of sediments with oil sands material. A pattern of toxicity was also observed, where the SP source material was the less toxic and ELLs material being more toxic. These results corresponded with the chemical analysis which showed the ELLs sample having high levels of PAHs and NAs. In summary, tests revealed that oil sands material affected model organisms under laboratory exposures, especially in samples with more NAs and PAHs content.

TU11 Genomic DNA methylation level : a stress molecular marker in the species Gammarus fossarum

Genotoxic evaluation has been developing for a couple decade among ecotoxicological assessment approaches in the aquatic field. It offers some prospect for understanding and predicting delayed effects on the offspring and the population dynamics (provided genetic mutations affect genetic genome). However, the modification of the DNA sequence by genetic mutation (as a result of primary DNA damage) is not the only impact of toxic substances on the genome. For example, epigenetic effects, defined as hereditary effects on the DNA function, may add up to mediated effects by genetics way. Among these marks, DNA methylation is extremely studied by scientists. It is so because it is a link to epigenetic changes in the context of population level perspective. This provides a complementary approach to effects on the primary structure of the genome for taking into account the time interval between the exposure to environmental contaminants and their effects. This issue is a real challenge in ecotoxicology. Moreover, epigenetic allows improving the understanding of changes of life history traits (reproduction, growth and development). These parameters are suitable for assessing the toxicity and are known to be regulated by epigenetic. Finally, epigenetic could also help better understand the variability of effects related to experimental conditions between the studies. Therefore, epigenetic marks have an innovative nature for the evaluation of genomic DNA methylation level as a possible stress biomarker in the ecologically relevant species Gammarus fossarum. First, the basal level was explored by studying the difference of overall DNA methylation between male, female and juvenile. Then, we evaluated the effects of natural factors on genomic DNA methylation level as starvation and exposed to different temperatures (8, 12 and 18°C) for 7 days and 1 month. To know the variability of DNA methylation in populations of Gammarus fossarum, three French populations from the same genetic strain, were studied. Lastly, the epigenetic biomarker response to a chemical stress was evaluated in a field experiment. We encaged gammarids from a reference unpolluted station in sites impacted by various human activities.

TU112 INVERTOX: Characterising individual metabolomic variability of the freshwater invertebrate, Gammarus pulex
T.H. Miller, Kings College London / Analytical and Environmental Sciences; J. MacRae, The Francis Crick Institute / Metabolonomics; N. Bary, University of Suffolk / Division of Diabetes and Nutritional Sciences; S. Owen, AstZeneca / Safety Health Environment; L. Barron, Kings College London / Analytical and Environmental Science.

The (pseudo)persistence of emerging contaminants and other organic micropollutants in the environment represents a risk for the organisms that are exposed to them. Metabolomics provides a powerful tool within environmental toxicology to better understand the impact of the exposure scenarios. However, metabolomics is an emerging field and to interpret the use of metabolite data for the understanding of toxicological responses is challenging. We need to know what is ‘normal’. The variability in individual metabolomes for a species, or a ‘background metabolome’ should be established to determine possible effects and the resilience of the population. This can only be done through influence data interpretation. Thus, we have characterised the effect of these factors on the metabolic variability in the freshwater invertebrate, G. pulex. Herein, an analytical method is presented for the extraction and non-target analysis1 of the metabolome in G. pulex. Briefly, a dual phase liquid extraction was used followed by HILIC-HRMS to enable detection and annotation of metabolic features extracted from individual animals. Animals collected from the field were analysed immediately and compared to animals that were extracted after a fixed period of acclimatisation to laboratory conditions. The results indicated that sex, mixing stage and acclimatisation period affected the metabolic variability and factors that are likely to influence metabolomic analyses should be investigated to aid understanding of pathways involved in effect-based studies. Furthermore, it may be possible to use selective analyses that are more focused to offer a better understanding in the data. Overall, the characterisation of metabolic variance for invertebrates along with the use of metabolomics shows a very powerful approach for understanding adverse effects that may be associated with environmental contaminants. References: 1) Zhang, T., et al. (2012). Analytical chemistry, 84(4), 1994-2001. Keywords: metabolomics; invertebrates; pharmaceuticals; modelling.

TU113 Ecotoxicological effects of the insecticide Imidacloprid on amphipods along pollution gradient in a river
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Many aquatic organisms are sensitive to man-made chemicals in the water. However, some species tolerate the occurrence of toxic chemicals and at the same time benefit from the nutrients that are often abundant in polluted waters. Such is also the case for amphipods of the species Gammarus pulex (Crustacea, amphipoda), which can be found throughout a pollution gradient of a stream. In our research, we investigated whether G. pulex individuals have an ability to adapt to the pollution gradient and to survive likewise in pristine and in polluted sites or whether different conditions select for different phenotypes within the population. G. pulex individuals were sampled at different sites along a pollution gradient in the river Holtemme (Saxony-Anhalt, Germany). Sites were characterized with respect to polluting burdens of water and amphipod tissues. Amphipods from three sampling points were brought alive to the lab and exposed to the insecticide Imidacloprid, which was found in water and animal samples beforehand. Lethal time for 50 % (LT50) values were determined and movement activities and moulting rates were recorded. Mortalities in the Imidacloprid treatments clearly differed for amphipods sampled at different sites; animals from more polluted sites were more sensitive to the toxicant and also exhibited reduced movement and moulting activities. We examined whether these differences in toxic sensitivities were related to differences in toxicokinetics of Imidacloprid by quantifying imidacloprid tissue levels after different times of exposure to the compound but toxicokinetics of imidacloprid proceeded similarly in animals from different sites. Population genetics approaches (sequence comparisons of a DNA stretch of the cytochrome oxidase I (COI) gene and comparisons of 9 microsatellite loci) revealed that differences in conditions between sites did not lead to the separation of distinct subpopulations suggesting that survival of individuals at each site is based on individual acclimation and not on adaptation to specific conditions by a distinct subpopulation.

TU114 Antenna Regeneration of the Marine Amphipod Parhyale hawaiensis as a Possible Endpoint in Ecotoxicology - Preliminary Data
O. Diehl, P. Assano, G. Umbuzeiro, School of Technology, UNICAMP / LAEG Parhyale hawaiensis is a marine amphipod of worldwide circumtropical distribution, which has been extensively studied as an ecotoxicological test species. P. hawaiensis is able to regenerate its appendages, limbs and tissues after an injury or lost during the entire course of their life. Regeneration can be used as an ecotoxicological endpoint to assess potential teratogenic compounds and their impact on stem cells. Studies suggest that P. hawaiensis has local progenitor cell in each part of body and it was already been demonstrated that P. hawaiensis has a fast regeneration of thoracic limbs, within a week, but no information on antennae’s regeneration was found. Thus, the aim of this study was to obtain data on regeneration of antennae of P. hawaiensis to determine the viability this endpoint on toxicity tests. On day one left antennae of six months old organisms were amputated with sterilized tweezers, each
organism transferred to recipients containing 100 mL salt water and a picture of each organism was taken under a stereomicroscope. Each test consisted of 20 organisms, 10 males and 10 females. During this period, organisms were fed three times a week, the necessary conditions of salinity, temperature, aeration, substrate and luminosity were provided. Four independent experiments were performed. The organisms were monitored daily until all of them undertook full regeneration. At the end of each test, another pictorial record was taken to determine the difference between the length (mm) before and after full regeneration. An average regeneration occurred from 7 to 20 days (n=80) after amputation and males and females behaved differently, Males took more time than females to complete regeneration. Length of the regenerated antennae varied from 50 to 80% of the original appendages to both male and female. Next steps will be the exposure of organisms to select toxicants to determine their ability in the regeneration process in the developed experimental conditions. Acknowledgement: Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq-PVE Process: 400362/2014-7) for funding and PIBIC for undergrad fellowship. Amanda dos Santos e Gabriel Rampazzo Magalhães for technical contribution.

TU115
Added value of community approaches in environmental risk assessment
M. Hammers-Wirtz, T. Strauss, Research Institute gaia / gaia - Research Institute for Ecosystem Analysis and Assessment; A. Toschki, Research Institute gaiae Community studies are an ecologically relevant tool to assess effects of stressors on population and community level. With these kinds of studies direct as well as indirect effects is a screening test addressed in lower tier testing to get a broader idea about the relevant effects on ecosystem structure and function; 2) as monitoring tool for products which passed risk assessment to check up on community level effects; 3) as monitoring tool for typical sequence scenarios of different products which will be used together in one crop. Here exemplary results of community studies and a screening study will be presented.

TU116
Metal pollution and macro-invertebrate communities in the Olifants River, Western Cape, South Africa
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Freshwater ecosystems are considered among the most threatened, as a result of current trends in water utilization globally. Within the Western Cape, 76% of rivers are polluted and run the risk of irreversibly losing their ability to support ecosystems. The developed test system, contr...
before test start. In a first step the test conditions were adapted to the requirements of mayfly larvae. Therefore testing of different media was performed. It turned out that, contrary to testing of stonefly larvae, which is performed in Cu-reduced dilution water, moderately hard reconstituted water (according to EPA) works best for testing of mayfly larvae. Instead of Tetramin®, which was used in stonefly testing, the green algae Desmodesmus subspicatus was used for feeding of mayfly larvae. The silvered mayfly larvae were acclimated for 48 hours, the acclimation period was extended to seven days before test start. Under these conditions mayfly larvae showed an acceptable mortality of test organisms. The next step is to perform a test with the test substance Imidacloprid which will be exposed to mayfly larvae for 21 days. Afterwards sensitivity of mayfly and stonefly larvae to Imidacloprid will be compared. The new testing method can provide toxicity data of different aquatic insects with different aquatic conditions, which can be used for a SSD (Species Sensitivity Distribution) approach. The developed test system, the results of the performed tests as well as a comparison between testing of mayfly and stonefly species will be presented.

TU119 Toxicity effects of a carbamate insecticide on a non-target freshwater gastropod: active ingredient versus commercial formulation

Carbamate insecticides are commonly used in agriculture for crop protection exerting their toxicity through the inhibition of the enzyme acetylcholinesterase. In Argentina, the maximum concentration of carbaryl (CAR) detected in surface and subsurface drainages was 45.7 µg L⁻¹. In this study, we evaluated the subchronic toxicity of environmental concentrations of the active compound and a commercial formulation of CAR on biochemical and reproductive parameters in Biomphalaria straminea, a freshwater gastropod native to Argentina. Five treatments were included in this study: dechlorinated tap water, acetone in dechlorinated tap water (solvent control), CAR active compound (dissolved in acetone) in dechlorinated tap water at 12.68 and 12.68 L⁻¹, and the equivalent to 12.68 L⁻¹ CAR of a formulation (dissolved in dechlorinated tap water) with 85% of the active compound. The concentrations used were chosen so as to have the same molarity as azinphos-methyl, an insecticide previously used in our laboratory. In bioassay 1, eight glass vessels per treatment were used with six snails each. After 14 days of exposure, homogenates were made with the organisms’ soft tissues (pool of five snails per vessel). In the supernatant fraction, the following parameters were measured: cholinesterases (ChE), carboxylesterases (CEs) with two substrates, glutathione S-transferase (GST), glutathione (GSH), superoxide dismutase (SOD) and catalase (CAT). In bioassay 2, bioassays, CAR solutions were renewed every 48 h based on previous stability studies. The active compound caused an increase in the activity of SOD with both treatment concentrations, whereas CAT activity was not affected by the treatment. Considering the previous findings, our results show that a subchronic exposure to CAR is toxic to the studied gastropod in our experimental conditions. Furthermore, with the 3Rs approach (reduction, refinement and replacement) the basic idea is to reduce the use of vertebrate organisms and to refine the procedures to minimize pain, suffering, and distress. To achieve this goal, ecotoxicology needs analytical tools, able to detect responses even at very low toxicant levels. At the CNR-ISMAR laboratory, it has been developed an innovative automatic recording system, namely Swimming Behavioral Recorder system (SBR system), coupled with an advanced image processing software. Nowadays, the SBR system has been used to record and track the swimming speed of different marine invertebrates, including crinidians, crustaceans, rotifers and echinoderms. In 10 years of research, SBR system has proved to be sensitive to a wide range of contaminants, such as metals, organic compounds, micro and nanomaterials, both polymeric and not, and even environmental matrices such as sediment elutriates. The amount of robust and significant data produced supports the suitability of this methodology to be applied to aquatic invertebrates. Here we reported a brief summary of SBRs’s applications, which show the relevance, sensitivity and versatility of the swimming speed alteration of marine invertebrates as ecotoxicological endpoint.

TU122 Benefits of Using Ecologically and Economically Valuated Invertebrate Species for Ecotoxicological Analyses: Potential Phototoxic Effects Comparing a Freshwater Vertebrate and Invertebrate
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The SBR system has proved to be sensitive to a wide range of contaminants, thus its use is increasing for ecotoxicological analyses in areas such as Louisiana where they are of such high importance. The use of crayfish, or other valued invertebrates, in ecotoxicology testing are additionally beneficial as they do not require IACUC approval and can likely be spawned in labs.

TU123 Impacts of anti-cancer drugs on freshwater rotifers at environmentally realistic concentrations
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Biomagnification in corals can occur through both filter and suspension feeding. It is a conundrum to classify uptake mechanisms because of the high cytotoxicity and increasing usage. The administration of drugs in coral, instead of single drug treatment, can make the assessment of the environmental risk of these compounds a difficult task with much information lacking on sub-lethal effects on aquatic species. We used two cytotoxic drugs aiming at linking their effects on the reproduction inhibition of the rotifer Brachionus calyciflorus with processes of oxidative stress. The rotifer was exposed to 0.1, 1, and 10 μM of a non-antibactericidal (5-Fluorouracil, 5FU) and a cytotoxic antibiotic (Doxorubicin; DOX) alone and in mixtures. The results showed that 5-Fluorouracil had a stronger effect (EC50=0.074 mg L-1) on the population growth rate than Doxorubicin (EC50=13 mg L-1) and toxicity effects were detected at environmentally relevant concentrations. Two concentrations of each drug were chosen for binary mixtures and two concentrations per drug were used to assess reactive oxygen species (ROS) accumulation and plasma membrane damage with epifluorescence microscopy. In the presence of low concentrations of SFU, there was a reduction of the toxicity induced by DOX indicating possible antagonistic effects between both drugs. At concentrations, as low as EC50, we found accumulation of ROS in a dose dependent manner showing a clear connection between ROS accumulation and the toxicity of these compounds. Furthermore, this indicates that both drugs not only have an effect on reproduction, cellular effects were found with possible consequences for the community at the long term.

TU124 Development in vitro and in vivo methods of measuring acetylcholinesterase and general esterases in aquatic invertebrates

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Quantification of acetylcholinesterase (AChE) and other esterases activities are important in terms of assessing the toxic mechanism of organophosphate and carbamate insecticides. There are several techniques for measuring AChE and general esterases (GE) activity involving spectrophotometric or fluorescence detection of transformation products. In this study, we tested four methods to detect AChE and GE activity in vitro and in vivo in the two aquatic invertebrate species: Daphnia magna and Chironomus riparius. The aim of this comparison was to study the efficiency and selectivity of the four methods. 2) to compare in vitro with in vivo measurements and 3) to compare the inherent esterase activities of D. magna and C. riparius. The four assays were: 1) AChE-assay using acetylthiocholine iodide (ATCI) as substrate, 5,5’-dithio-bis-(2-nitrobenzoic acid) (DTNB) as chromogen, measuring the production of 5-thio-2-nitrobenzoic acid; 2) AChE-assay using acetylcholine (ACh) as substrate, measuring resorufin formation; 3) GE-assay using 1-naphthyl acetate (1-NA) as the substrate, measuring 1-naphthol production and 4) GE-assay using 4-methylumbelliferyl butyrate (4-MUB) as the substrate, measuring 4-methylumbellifereone production. Michaelis-Menten curves were created for all substrates, where it was possible. The results showed that the GE-assay using 4-MUB measured general esterase activities well both in vitro and in vivo in C. riparius. However, the AChE and ATCI methods with 24-h preparations could only be used in vitro, while the AChE-assay using resorufin formation could not be used either in vitro or in vivo. The maximal GE-activities in vitro in D.magna and C. riparius were 345±44 and 151±51 nmol min-1 mg protein-1 respectively, when using 1-NA and 295±8 and 60±13 when using 4-MUB, hence, showing comparable activities across substrates. Focusing only on AChE-activity in vitro the maximal activities were 13.2±0.3 and 52.3±1.1 nmol min-1 mg protein-1 in D.magna and C. riparius, respectively, making C. riparius the species with the highest activity. The turning in to in vivo measurements, the GE-activities were 49.1and 17.4 nmol min-1 mg protein-1 for D.magna and C. riparius. The results of GE-assays using 1-NA and 4-MUB are similar. The AChE-assay could not be conducted in vivo. The GE-assay using 4-MUB, however, could be conducted in vitro as well as in vivo. The GE-activity in D.magna was higher while the AChE-activity in D.magna was lower compared to C. riparius.

TU125 Factors influencing bioaccumulation of metals and pollutants in corals

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As human population increases, the presence of emergent chemical contaminants (ECCs) in freshwaters increases. ECCs have shown to be persistent and bioactive, reaching the freshwater aquatic systems mostly untreated, where their fate and behavior is little understood. Ant-cancer drugs are among the ECCs of concern due to their high cytotoxicity and increasing usage. The administration of drugs in corals, instead of single drug treatment, make the assessment of the environmental risk of these compounds a difficult task with much information lacking on sub-lethal effects on aquatic species. We used two cytotoxic drugs aiming at linking their effects on the reproduction inhibition of the rotifer Brachionus calyciflorus with processes of oxidative stress. The rotifer was exposed to 0.1, 1, and 10 μM of a non-antibactericidal (5-Fluorouracil, 5FU) and a cytotoxic antibiotic (Doxorubicin; DOX) alone and in mixtures. The results showed that 5-Fluorouracil had a stronger effect (EC50=0.074 mg L-1) on the population growth rate than Doxorubicin (EC50=13 mg L-1) and toxicity effects were detected at environmentally relevant concentrations. Two concentrations of each drug were chosen for binary mixtures and two concentrations per drug were used to assess reactive oxygen species (ROS) accumulation and plasma membrane damage with epifluorescence microscopy. In the presence of low concentrations of SFU, there was a reduction of the toxicity induced by DOX indicating possible antagonistic effects between both drugs. At concentrations, as low as EC50, we found accumulation of ROS in a dose dependent manner showing a clear connection between ROS accumulation and the toxicity of these compounds. Furthermore, this indicates that both drugs not only have an effect on reproduction, cellular effects were found with possible consequences for the community at the long term.
the cell, they require a multistep metabolic activation by specific enzymes that participate in biotransformation reactions. The aim of this study was to evaluate biochemical and molecular biotransformation responses of the oyster *Crassostrea brasiliana* exposed to pyrene (50 mg L\(^{-1}\) and 100 mg L\(^{-1}\)) and fluorene (100 mg L\(^{-1}\) and 200 mg L\(^{-1}\)), after two time periods of exposure (24 h and 96 h). The half-life times of both PAHs were quantified by fluorescence in the aquaria exposure water and the transcription of phase I (CYP1-like, CYP2-like, CYP2A1U and CYP156A1-like) and phase II (GST-like, GSTm-like and SULT-like) biotransformation genes, EROD, GST and GSTm activity, were evaluated in gills. The half-life time of pyrene (100 mg L\(^{-1}\) = 2 h and 12 min) in water was lower than fluorene (100 mg L\(^{-1}\) = 5 h and 54 min). These results might be related to the higher lipophilicity of pyrene, facilitating its influx through the plasma membrane into the intracellular compartment and enzymes in pyrene biotransformation. This study contributes to the identification of new biomarkers of PAHs contamination in *C. brasiliana*. Also evidences a possible participation of these genes and enzymes in pyrene biotransformation metabolism. In addition, it suggests the participation of CYP2A1U gene in the biotransformation process of PAHs in gills of *C. brasiliana*. 

**TU128**

**BIOCHEMICAL AND CELLULAR RESPONSES OF THE CRAB *PACHYGRUS MARMORATUS* TO EVALUATE THE ENVIRONMENTAL CONTAMINATION OF THE LIVORNO HARBOUR (ITALY) AND OF AN ADJACENT MPA**

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The main purpose of the present investigation was to assess the toxicological status of Livorno harbour in Italy and of an adjacent MPA, through the responses of a set of biomarkers in the crab *Pachygrus marmoratus*. This investigation is part of the IMPACT project (Port on Impact on Marine Protected Area: cross-border co-operative actions), which has the purpose to design cross-boundaries management plans to actually protect the Marine Protected Areas. Male and female crabs were collected from the different areas: Livorno harbour, considered a polluted area, and the MPA protection area and environments of the MPA borders management. The crab *P. marmoratus*, one of the most significant indicator species to monitoring coastal marine environment, was chosen as model organism, able to provide information on the acute and chronic toxicity of potentially toxic compounds (metals, surfactants, pesticides, nano-particulates) and on their contribution to the global pollution. This species is used in several studies and offers a good opportunity to assess the effects of different contamination on stress-related biomarkers, metabolic capacity and energy reserves, after 96 hours exposure periods, were evaluated. The obtained results revealed significant alterations in contaminated mussels, varying with the concentration and time of exposure. Mussels exposed to Ti presented lower metabolism, represented by lower oxygen pulse and lower heart rate (Fp) of ephyrae, activity, which decreased along exposure time. The decrease of metabolic parameters were evaluated for the different exposure periods (24 and 96 h) and phase II (CYP1-like, CYP2A1U and CYP156A1-like) and phase II (GST-like, GSTm-like and SULT-like) biotransformation genes, EROD, GST and GSTm activity, were evaluated in gills. The half-life time of pyrene (100 mg L\(^{-1}\) = 2 h and 12 min) in water was lower than fluorene (100 mg L\(^{-1}\) = 5 h and 54 min). These results might be related to the higher lipophilicity of pyrene, facilitating its influx through the plasma membrane into the intracellular compartment and enzymes in pyrene biotransformation. This study contributes to the identification of new biomarkers of PAHs contamination in *C. brasiliana*. Also evidences a possible participation of these genes and enzymes in pyrene biotransformation metabolism. In addition, it suggests the participation of CYP2A1U gene in the biotransformation process of PAHs in gills of *C. brasiliana*. 

**TU129**

**Toxicity of titanium on the mussel *Mytilus galloprovincialis***

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Titanium (Ti) is at forefront of research related to nanomaterials. Due to their physical and chemical properties, Ti nanoparticles (TiO\(_2\)) are widely used in various industrial applications, such as additives in pharmaceuticals and food colorants, toothpastes, solar cells, sunscreens, cosmetics and boat paints. With the increasing production and use of TiO\(_2\), Ti has been inevitably released into aquatic systems through wastewater treatment plants, surface run-off, direct inputs and atmospheric deposition. The increasing input of TiO\(_2\) in the aquatic environment has raised concerns about the toxicity of Ti to inhabiting organisms. Once in the aquatic environment TiO\(_2\) interacts with the surrounding water components, including other contaminants, which may change the availability of Ti to organisms, namely the ability to penetrate into cells which may result in toxicity. In the present study the mussel species *Mytilus galloprovincialis* was used to evaluate the impacts caused by the exposure of Ti (II) solutions with the initial concentrations of 5pg/L, 50pg/L, 100 pg/L of Ti (II). Biochemical (oxidative stress related biomarkers, metabolic capacity and energy reserves) markers, after 96 hours and 14 days exposure periods, were evaluated. The obtained results revealed significant alterations in contaminated mussels, varying with the concentration and time of exposure. Mussels exposed to Ti presented lower metabolism, represented by lower oxygen pulse and lower heart rate (Fp) of ephyrae, activity, which decreased along exposure time. The decrease of metabolic parameters were evaluated for the different exposure periods (24 and 96 h) and phase II (CYP1-like, CYP2A1U and CYP156A1-like) and phase II (GST-like, GSTm-like and SULT-like) biotransformation genes, EROD, GST and GSTm activity, were evaluated in gills. The half-life time of pyrene (100 mg L\(^{-1}\) = 2 h and 12 min) in water was lower than fluorene (100 mg L\(^{-1}\) = 5 h and 54 min). These results might be related to the higher lipophilicity of pyrene, facilitating its influx through the plasma membrane into the intracellular compartment and enzymes in pyrene biotransformation. This study contributes to the identification of new biomarkers of PAHs contamination in *C. brasiliana*. Also evidences a possible participation of these genes and enzymes in pyrene biotransformation metabolism. In addition, it suggests the participation of CYP2A1U gene in the biotransformation process of PAHs in gills of *C. brasiliana*. 

**TU130**

**Comparing interspecific Artemia responses to chronic zinc exposure**

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The invasive species *Artemia franciscana* is displacing native Artemia (*A. salina* and *A. parthenogenetica*) from eastern Atlantic coasts and across the Mediterranean region, where they are used as food for native Artemia species when exposed to zinc. However, recent studies suggest that local adaptation to the contaminated conditions by the native population may prevent colonization by *A. franciscana*. Under this context, the sublethal toxicity of zinc was assessed in natural populations of *A. parthenogenetica* from the highly contaminated Odile estuary (southern Spain) and of *A. franciscana* from the less contaminated one (Cadiz bay, SW Spain). The Zn concentration used in our experiments (0.2 mg/L) was the double of that recorded in water from the Odile estuary to make our results as relevant as possible to real field conditions. Cysts were hatched in seawater and nauplii (*A. parthenogenetica*) or separated in couples (*A. franciscana*) according to their group (control and treatment) and a set of reproductive parameters were examined. Results showed that *A. franciscana* performs better (higher survival and growth) than *A. parthenogenetica*. Both species experienced significant slower growth and higher mortality when exposed to Zn, but non significant effects were found in final size. Regrading reproductive parameters, Zn exposure increased offspring production of both Artemia species when compared to control. *A. franciscana* displayed a better performance (higher number of broods and offspring production; lower % non-viable nauplii) than *A. franciscana*. The results of this work highlight the competitive advantages of native species (*A. parthenogenetica*) from contaminated areas to prevail under the selective pressure of abiotic factors as environmental pollution. Based on these results the highly polluted Odile estuary would not be a relevant habitat for native Artemia species when compared to the more polluted one. The study of *A. franciscana* as a model organism has interesting applications in the field of ecotoxicology, especially in the study of the effects of pollutants on the reproduction of aquatic species. 

**TU131**

**Promising invertebrate species as model organism in ecotoxicology: ephyrae of the jellyfish Aurelia sp. and Sanderia malayensis**

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In aquatic toxicology, one of the most important steps is the selection of suitable model organisms, able to provide information on the acute and chronic toxicity of marine pollutants. In this context, invertebrates species are being used extensively in laboratory tests for their usefulness for seeking mechanistic links between effects occurring at the individual level and consequences for higher levels of biological organization. In addition, compared to vertebrates they are also easy to maintain under laboratory conditions, widely distributed and ecologically relevant. Although Cnidarian jellyfish (Scyphozoa) are known to play an important role in marine food webs and are often used as model organisms in ecotoxicological test, they are not yet employed in routine ecotoxicology. The aim of this current investigation is to suggest the use of two new invertebrate species of the jellyfish *Aurelia sp.* and *Sanderia malayensis* as model organisms in ecotoxicological bioassays. A series of experiments were carried out in laboratory controlled conditions, in order to characterize some experimental parameters that can influence the Frequency of a given endpoint, as the end points of the experiments. The results are a set of sub-lethal response for this innovative invertebrate model for ecotoxicological testing. After these preliminary tests, ephyra were exposed to a wide range of potentially toxic compounds (metals, surfactants, pesticides, nano-materials and harmful algae, emerging compounds), in order to evaluate the potential of ephyra jellyfish in ecotoxicology. The experiments allowed to identify two end-points (sub-lethal, frequency of pulsation and acute, immobilization) with different levels of sensitivity and to optimize the use of an automatic recording system of swimming marine invertebrates (Swimming Behavioural Recorded e SBR), already employed with other biological models. In addition, the comparison of the ECso
values obtained exposing ephyrae jellyfish to different toxic compounds and materials such as nanoparticles and microplastics with those obtained with other marine invertebrates, highlights that ephyrae are an interesting and promising invertebrate model with a very high ecological relevance to be used in ecotoxicological investigations.

TU132 Paracentrotus lividus and Artemia sp.: never too old model organisms to give new end-points
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In the last few years it has become increasingly important the contribution of ecotoxicological assays to the environmental monitoring, as a fundamental instrument of chemical analyses. In environmental risk assessment, in order to fulfill several regulatory requirements, such as the 3R principles (reduction, refinement and replacement), the development of novel approaches to reduce and eventually substitute the use of vertebrate species results to be paramount.

Swimming alteration is one of the most frequently used behavioral responses in aquatic ecotoxicology and its evaluation has proved to be a valuable endpoint in ecotoxicological studies with aquatic organisms. Behavioral responses have proven their usefulness in evidencing impacts of chemicals at environmental concentration that do not necessarily cause mortality; therefore, behavioral endpoints are less invasive than traditional acute tests, but still sensitive and more ecological relevant.

In this work, we reported a novel research on the use of swimming behavior of two “old” marine model invertebrates in ecotoxicology, the crustacean Artemia sp. and the echinoderm Paracentrotus lividus, concentration dependent swimming alteration test (SDST) to lead to an endpoint. In detail, we optimized and improved an automatic recording system, namely Swimming Behavioral Recorder system (SBR), by developing i) a new swimming speed alteration test using for the first time sea urchin early stages; ii) a new short-term test based on the evaluation of the swimming speed alteration of Artemia nauplii incubated at 39 °C (± 1) for only 6 hours. Thanks to a modern video-based technology, this study provides novel perspectives and future applications applied to two well-known marine model invertebrates, meeting regulatory and market demands, including the reduction in using vertebrate species and the need for early warning technologies.

TU133 Application of sea-urchin embryo test in the effect directed analysis approach for the evaluation of WWTP effluent in an estuarine media
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Since target chemical analysis often cannot explain the cause-effect relationship between certain contaminants and the observed effects in organisms, effect directed analysis (EDA) can be applied to unravel the drivers of toxicity in complex mixtures. In this study, the sea-urchin embryo test (SET) was implemented for the first time in a EDA approach in order to evaluate an estuarine environment influenced by the effluent of the main waste-water treatment plant (WWTP) of Bilbao (WWTP Bilbao). The effluent was extracted by a sequential LC-UV fractionation methodology based on two different columns: a Nucleobond C8 column (21 fractions were collected) and an aminopropyl column (15 fractions). Two endpoints were used to detect the toxic effects after 48 h: the growth rate of the larvae and the rate of skeletal malformation. 6 levels (n=3) of dose-curve were prepared in units of relative enrichment factor (REF, final volume of 3 ml of filtered seawater with 0.1 % of DMSO). Non-target analysis was performed by means of UHPLC-Qxactive Plus MS in positive and negative modes with a C18 column. Toxic compounds were identified using MS2 spectra, Metfrag and Compound Discoverer (Thermo) interfaced to MZmine. Among the collected C8 fractions, only fraction 113 (F113) showed a clear toxicity and, therefore, it was tested separately to establish the concentration-response model. The curve-dose response of the raw sample (EC50 = 10 REF and EC25=19 REF) could be explained by the contribution of active F13 (EC50=14 REF and EC25=39 REF).

Regarding the chemical analysis, among the final candidate list (20 compounds), mendazolone (an anilimethinic agent) was confirmed chromatographically with standards. Nevertheless, a sequential fractionation of F113 was also carried out with an aminopropyl column, which showed a different orthogonality compared to C8 column, and the resulting 15 fractions were also submitted for further bioassays and data-dependent analysis. Overall, the results of this work suggest the possibility of addressing a kind of specific toxicity in sea-urchin embryos owing to the determination of only one toxic fraction and the contaminants identified in that fraction. Acknowledgement. This work was financially supported by the Ministry of Economy and Competitiveness through the project CTM2014-56628-C3-1-R. L. Mijangos is grateful to the Basque Government and H. Ziarrusta to the Spanish Ministry for their predoctoral fellowships.

TU134 Plausibility of Daphnia magna model to evaluate eicosanoid pathway related toxicity
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Eicosanoids are biologically active, oxygenated metabolites of C20 polyunsaturated fatty acids and are synthesized through cyclooxygenase, lipoxigenase or cytochrome P450 epoxide pathway. As signaling molecules, they are important for diverse physiological systems such as inflammation, allergy, pregnancy, pain perception and blood pressure control. Therefore, they could be the important target for toxicants or drugs such as ibuprofen and aspirin. However, invertebrate model to evaluate eicosanoid pathway related toxicity has not yet been developed well though similar eicosanoid pathway was expected in invertebrate species. Therefore, the purpose of this study was to identify genetic information of key genes of eicosanoid pathway in Daphnia magna, a widely used invertebrate model, and to evaluate the transcriptional levels of the genes after the exposure to eicosanoid targeted drugs. To this end, we collected amino acid sequences of eicosanoids from other species and then compared the sequences in water flea genome database. After the BLAST and alignment, the genetic information of 10 key eicosanoid genes, such as pla2, cox, pgd2a and pge2, was identified. After that, Daphnia magna was exposed to the eicosanoid pathway targeted drugs, i.e., ibuprofen, indomethacin, celecoxib and acetaminophen at 0.25, 2.5 and 25 μM. Then, we analyzed transcriptomes. To ensure accuracy, we performed a chronic multi-generation exposure (nine generations) to lead (0.05 mg/L Pb) under different dietary exposures (regular and restricted) and, regarding chemical mixtures of natural environments, pulse exposures (on Pb acclimated daphnids) to the fungicide mancozeb were also accomplished. Organisms from Pb were changed to a clean media for recovery for three generations. To monitor acclimation, standard acute immobilization tests to K+C6O4- (to check for sensitivity), to the metal Pb and to mancozeb were made. Since size is crucial on organisms’ tolerance, neonates’ body length was also measured. No difference on daphnids sensitivity was spotted among generations, except D. similis from recovery period under food restriction. However, Pb tolerance increase is seen on both species. Regarding mancozeb exposure (Pb acclimated), no difference between treatments is seen under regular food. However, opposite outcomes are shown under food restriction, such as D. magna increasing and D. similis decreasing tolerance to mancozeb. Adverse outcomes regarding recovery was shown, D. magna relied on genetic adaptation, since it kept a higher tolerance to Pb, even after three generations under recovery, and D. similis relied on physiological acclimation, being similar to organisms from control treatment. Food restriction also decreased reproduction and increased neonates’ body length (both species). The opposite outcomes under food restriction is crucial regarding natural environments and the natural fluctuating amount of nutrients. Therefore, results indicate that daphnids are capable of acquiring resistance to Pb under a long-term exposure, being an essential data on chemical mixtures contaminated habitats, since their tolerance to other chemicals can vary (as shown for mancozeb). And also, opposite outcomes regarding monophyletic species indicates that it is not accurate to use species from different climates to estimate toxicity.

TU135 Responses to single chemical and pulse exposures of two monophyletic Daphnia species under a multi-generation approach
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Phytoplankton human activities are continually exposed to environmental contamination (e.g. industries, agriculture). Those contaminants may have continuous or pulse sources and can affect organisms from natural habitats. In different latitudes even phylogenetically close related species may present divergent chemical tolerance. Therefore, it was used in this study the model species from temperate areas Daphnia magna and the tropical species Daphnia similis. Most studies rely on short-term acute and chronic tests. To ensure accuracy, we performed a chronic multi-generation exposure (nine generations) to lead (0.05 mg/L Pb) under different dietary exposures (regular and restricted) and, regarding chemical mixtures of natural environments, pulse exposures (on Pb acclimated daphnids) to the fungicide mancozeb were also accomplished. Organisms from Pb were changed to a clean media for recovery for three generations. To monitor acclimation, standard acute immobilization tests to K+ C6O4- (to check for sensitivity), to the metal Pb and to mancozeb were made. Since size is crucial on organisms’ tolerance, neonates’ body length was also measured. No difference on daphnids sensitivity was spotted among generations, except D. similis from recovery period under food restriction. However, Pb tolerance increase is seen on both species. Regarding mancozeb exposure (Pb acclimated), no difference between treatments is seen under regular food. However, opposite outcomes are shown under food restriction, such as D. magna increasing and D. similis decreasing tolerance to mancozeb. Adverse outcomes regarding recovery was shown, D. magna relied on genetic adaptation, since it kept a higher tolerance to Pb, even after three generations under recovery, and D. similis relied on physiological acclimation, being similar to organisms from control treatment. Food restriction also decreased reproduction and increased neonates’ body length (both species). The opposite outcomes under food restriction is crucial regarding natural environments and the natural fluctuating amount of nutrients. Therefore, results indicate that daphnids are capable of acquiring resistance to Pb under a long-term exposure, being an essential data on chemical mixtures contaminated habitats, since their tolerance to other chemicals can vary (as shown for mancozeb). And also, opposite outcomes regarding monophyletic species indicates that it is not accurate to use species from different climates to estimate toxicity.

TU136 Chronic effects of BPA, BPS, and BPSIP in Daphnia magna
Y. Hong, B. Jeon, I. Ryoo, J. Lee, K. Ji, Yongin University
Since bisphenol A (BPA) can act as a weak estrogen agonist and disrupt steroidogenesis, concerns on adverse health effects are increasing. In response to the regulatory pressures to eliminate BPA in plastics, bisphenol S (BPS) and...
4-hydroxyphenyl 4-isoproxyphenylsulfone (BPSIP) have been frequently used in manufacture of thermal paper and plastic containers. However, there is paucity of information on their chronic toxicity using aquatic invertebrates. In the present study, chronic toxicity of BPA, BPS, and BPSIP were evaluated using Daphnia magna in accordance with OECD Test Guideline 211. The endpoints for the long-term exposure were survival, reproduction, and growth. Compared to the control group, the body length was significantly decreased in D. magna exposed to 1 mg/L of BPA, 5 and 10 mg/L of BPS, and 1 mg/L of BPSIP (<0.05). The results showed that BPA, BPS, and BPSIP could induce endocrine disruption related to the growth in aquatic invertebrates, and the effective concentration of BPSIP was similar to that of BPA. With increasing use of these alternative compounds, more monitoring program in aquatic environment and study of toxicity mechanism appears to be necessary. Acknowledgement: This study was supported by the National Research Foundation of Korea (Project NRF-2015R1D1A1A01056628).

TU137 Oxidative effects of mono-(2-ethylhexyl)-phthalate on Daphnia magna in both molecular and population level
Y. Kim, KIST Environmental Safety Group; Y. Seo, KIST-Europe / Environmen
safety group
Mono-(2-ethylhexyl)-phthalate (MEHP) is the metabolite of di-(2-ethylhexyl)-phthalate (DEHP), which is widely used in the industry as plasticizers. According to previous studies, DEHP inhibits molting hormone. In addition, MEHP is highly persistent and bioaccumulative in environment and living organisms. In EU Regulation 2008/105/EC on plan of action for MEHP acts as an estrogen disrupting chemical in aquatic organism such as Daphnia Magna. The aim of this study was to elucidate the linkages between toxicity test result and oxidative stress of MEHP. We studied the effects of oxidative stress as molecular initiating events on Daphnia magna. We observed the changes in different levels of the lipid peroxidation, glutathione S-transferases (GSTs), catalase (CAT) and superoxide dismutase (SOD) in the treated daphnids. This study showed the molting rate, reproduction rate and growth of daphnids during chronic (21 d) test in order to link the activities of reproduction system to antioxidant responses. Further study is needed to clarify how MEHP leads to dysfunction of endocrine system of Daphnia magna.

TU138 Are Daphnia magna and Chironomus riparius acute responses comparable? B. Ponti, ChemService Srl; R. Bettinetti, University of Insubria / DISTA; D. Garagna, ChemService s.r.l.; F. Casarotto, University of Insubria; M. Neri, ChemService srl - Controlli e Ricerche
Laboratory ecotoxicity test results predict the responses of organisms with varying degrees of accuracy. Traditionally, the acute toxicity on aquatic invertebrates is estimated by exposing for 48 hours young cladocerans of Daphnia magna (OECD test guideline n. 202, 2004), taking advantage of its well-experienced sensitivity and reliability for a huge number of known and unknown toxicants. The 48 hours test on Daphnia magna conducted according to OECD 202 is listed as a data requirement in EU Regulation 2008/105/EC on plan of action for MEHP as well as in Reg EC 1907/2006 on chemicals (REACH). In recent years even the use of first instar larvae of Chironomus riparius (Insecta, Diptera) has been proposed (OECD test guideline n. 235, 2011) to be used to complement existing Test Guidelines for chironomid chronic toxicity assays (OECD test guidelines 218, 219, 2004) (OECD guideline n. 235, 2011) to be used to complement existing Test Guidelines for chironomid chronic toxicity assays (OECD test guidelines 218, 219, 2004) (OECD guideline n. 235, 2011) to be used to complement existing Test Guidelines for chironomid chronic toxicity assays (OECD test guidelines 218, 219, 2004). The aim of this work was to assess genetic variation in tolerance in the aquatic insect Chironomus riparius exposed to microbial insecticides. A C. riparius population was established in the laboratory by crossbreeding five populations, in order to ensure sufficient levels of genetic diversity. By employing a full-sib family split design, this study presents a quantitative genetic analysis among families of C. riparius across different environments (microbial insecticide exposure under two levels of salinity). Each egg mass was considered a single family (full-sib genotype). Within each family, larvae were randomly allocated to all treatments. Emergence rate, time to complete development, mean biomass weight were used as response variables. Relationships between genetic variation, life-history traits and fitness costs in response to microbial insecticides, and the potential of a key aquatic insect species to evolve tolerance to these compounds will be discussed as well as an evolution of disease resistance on aquatic species. The study also highlights the suitability of C. riparius, a model organism in aquatic toxicology, for quantitative genetic analyses.

TU140 Genetic variability in tolerance to microbial insecticides in Chironomus riparius
M. D. Bordalo, University of Aveiro; A. Rodrigues, University of Aveiro / Biology Department; and S. Pestana, Cesam & University of Aveiro / Biology Department; I. Lopes, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro; J. Pestana, CESAM & University of Aveiro / Biology Natural populations are constantly facing a large array of environmental stressors, from both natural and anthropogenic origin, which represent a strong selective force shaping the behaviour, physiology and morphology of organisms. Genetic diversity acts as a key component of adaptation to biotic and abiotic stressors. However, there is paucity of information on their chronic toxicity using aquatic invertebrates. In the present study, chronic toxicity of BPA, BPS, and BPSIP were evaluated using Daphnia magna and Chironomus riparius in both molecular and population level. These mixtures can reach the biota of freshwater ecosystem so the main objective was studying the effects of the mixtures that can reach on a invertebrate with a relevant role in the food chain of these ecosystems, Chironomus riparius. C. riparius is a dipteran with aquatic larvae frequently used in toxicity tests. Fourth instar larvae were exposed for 24 hours to single compounds and to binary and ternary mixture of MEHP as well as BPA by retrotranscription and Real-Time PCR using a specific array covering a relevant number of metabolic pathways like endocrine system, immune response, stress response, detoxification mechanisms and apoptosis among others. Using an array could improve the toxicological evaluation of the cellular effects of the compounds favoring the identification of new molecular biomarkers useful for ecological risk assessment and toxicity tests. The methodology used to design the array can be used with other species improving also our knowledge about the mode of action of these compounds. This work has been funded by the Ministerio de Economía y Competitividad, CIICYT (SPAIN), CMT2015-64913-R/a.a.B.M.G.M is the receiver of a pre-doctoral contract from the National University of Distance Education (UNED).

TU141 Effects of Amitraz on Chironomus riparius: life history and biochemical responses
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Amitraz is a very effective formamidine insecticide used in agriculture to control fruit trees and cotton pests. Due to its widespread use and high persistence, there is an elevated risk of contamination of adjacent aquatic ecosystems. However, the information of amitraz toxicity to aquatic non-target invertebrates is scarce. In this study, the toxicity of amitraz to the midge Chironomus riparius (Meigen) was assessed in terms of life history responses using standard ecotoxicological tests and biochemical endpoints using oxidative damage, antioxidant defenses, energy metabolism, and neuromuscular toxicity biomarkers. Chronic exposure to amitraz contaminated waters (28 days; 10, 20, 40, 80, and 160 µg L⁻¹) resulted in impairment of C. riparius emergence and developmental rates (reduced larval growth and delayed emergence), with male development time being
the most sensible endpoint (LOEC of 40 µg L⁻¹). Short-term exposures (48 h; 0, 10, 40, and 160 µg L⁻¹) to amitraz induced glutathione peroxidase activity and a decrease in catalase activity. Additionally, amitraz exposure caused a decrease in lactate dehydrogenase activity and a significant increase in electron transport system activity, both energy metabolism associated biomarkers. Regarding oxidative damage biomarkers, lipid peroxidation increased in C. riparius larvae exposed to 100 µg L⁻¹ amitraz, while there was a significant decrease in DNA damage levels at 10 and 40 µg L⁻¹ treatments. The study results reveal possible biochemical targets of amitraz toxicity and suborganismal responses associated with amitraz exposure. The assessment of biochemical biomarkers may support the interpretation of toxic responses observed at organism level and therefore in the assessment of the ecological effects of environmental contamination. Moreover, it increases the information available on the outcomes of amitraz exposure in freshwater invertebrates, and underlines the importance of risk assessment studies of formamidine pesticides. Acknowledgements: This study had the support of the Fundação para a Ciência e a Tecnologia through project PROTEOME (PTDC/AAG-MAA/1302/2014), co-financed by COMPETE (POCI-01-0145-FEDER-16773).

TU142 Multigenerational exposure of Folsomia candida to copper agrochemicals: conventional and nano-pesticides

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Biotechnological and Animal Cell Biology, Research Centre for Experimental Marine Biology and Experimental Interaction of the Environment, University of Aveiro / Biology; S. Loureiro, Universidade de Aveiro / Biology

Agricultural practices include the use of agrochemicals for crop maintenance and enhanced production. Although soil contamination may result in long-term ecotoxicological effects, like copper pesticides, have been used in a range of agricultural applications, which may result in environmental problems. More recently, nanopesticides were introduced in the market with the intent to improve efficacy and decrease environmental negative effects. However, the chronic ecotoxicological effects of nanopesticide exposure on soil biota are not well known since related environmental hazards are most often assessed using only the active ingredients. Moreover, the multigenerational effects of long-term, chronic exposure of soil organisms to agrochemical applications are unknown. The objective of this study was to evaluate the impact of long-term, multigenerational exposure of the soil collembola Folsomia candida to conventional and nanoparticle formulations of copper pesticides. Two formulations were assessed: Kocide® 3000 (nano form) and Champion® WP (conventional), as well as the pure active ingredient Cu(OH)₂ in spiked LUFA 2.2 soil. The effects of multigeneration exposure to the Cu pesticides were assessed using two soil treatments: 1) Cu spiking performed only at the beginning of the experiment and collombolan responses (survivorship, reproduction) measured for three generations (i.e., aging soil exposure); and, 2) Cu spiking performed at the start of each new cohort (i.e., repeated soil exposure). After three generations in both soil treatments, the surviving collombola were moved to uncontaminated soil for three generations to assess their recovery potential. Similar response patterns were observed in the two soil treatments for all three Cu formulations. Exposure to aging soils revealed an increasing tolerance across generations of F. candida. In contrast, in treatments with renewed Cu spiking, the collombolan populations showed ongoing sensitivity to Cu exposure. In both treatments, after being moved to clean soil, all treatment populations showed some recovery by displaying increased reproductive output. Copper forms presented different behavior between them in the long term exposure. This study further emphasises the importance of using multigenerational approaches to obtain more ecological relevant evaluations of environmental risk associated with chronic exposure to soil agrochemicals.

TU143 Effects of multiple environmental stressors on Eisenia fetida coelomocytes: cell viability and different behaviour of amoebocytes and eleocytes

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Earthworm immune cells (coelomocytes) have become a target system in ecotoxicology due to their sensitivity against a wide range of pollutants. Moreover, endpoints measured in coelomocytes retrieved from exposed Eisenia fetida offer rapid and accurate information to predict impairments caused by pollutants at longer exposure times and higher complexity levels (organism, population). Since soils are subjected to multiple environmental stressors (i.e., temperature increases, acidification, organic matter depletion, new pollutants) it is of great interest to assess how those stress scenarios pose changes in earthworms at cellular level. Coelomocytes compose a heterogeneous cellular group where two major cell subpopulations are distinguished, amoebocytes and eleocytes. However, the behavior of those subpopulations against different stressors is still unclear. Hence, the aim of the present work was to address the effects of different stressors (increase in temperature, low OM and model and emerging –nanoparticles- contaminants) on E. fetida coelomocytes by assessing mortality and changes in the relative proportion of subpopulations (amoebocytes, eleocytes). For that, earthworms were maintained under low OM content (6% vs. 10%), thermal stress (19ºC vs 26ºC) and new conditions (Cd: 5-25 mg kg⁻¹ dw.; Ag NPs: 0-100 mg kg⁻¹ dw) in OECD soil during 3 days. After exposure of earthworms, coelomocytes were retrieved and viability was assessed in microplate through Calcein AM assay. In addition, flow cytometry analysis was used to determine mortality of coelomocytes and changes in the relative proportion amoebocytes/eleocytes. Coelomocytes extruded from earthworms maintained at low OM content, higher temperatures showed a decrease in cell viability, but no changes were recorded in the relative proportion of amoebocytes and eleocytes. Exposure to Cd provoked higher mortality in eleocytes while Ag NPs caused more mortality in amoebocytes. Thus, we can conclude that the response of the different subpopulations was dependant on metal form and was enhanced by environmental factors (increased temperature and low OM). These results reinforce the potential role of coelomocytes as an accurate soil health assessment in a global warning scenario. Acknowledgements: Basques Gov. (IT810-13), Univ. Basque Country (UIF 11/37) and MINECO (Nanosilveromics Project).

TU145 Terrestrial arthropods as indicators of environmental pollution

V. Leach, North-West University; H. Bouwman, North-West University / Unit for Environmental Science and Management

In recent years, the use of and interest in terrestrial arthropods as indicators of environmental pollution has increased. Arthropods are diverse, with over 31 000 species described and collection normally has less ethical restrictions then for higher animals. We reviewed the literature. We found relatively few general studies on arthropods as indicators of environmental pollution. However, those that did worked on spiders, bees, earthworms and ants. In addition, most studies favoured predatory species, since this the most sensitive group. We give insights into the potential use of arthropods in the area. In most studies, the sampling sites were close to old mines, or the studies were comparisons of arthropods from different locations. Published literature on terrestrial arthropods not only focuses on whole body utilization but also organ specific studies, as well as research on the use of arthropod products (such as honey and spider webs) as matrices for analyses. Most arthropods have a close association with soil, foliage and air, representing pollution concentrations in the immediate environment. Each arthropod species occupy a unique niche and in doing so represent the concentration or degree of pollution in different ways. Each species, therefore, indicates pollution from different perspectives. However, there is still a
TU146 The impact of chlorpyrifos and its formulations on the acetylcholinesterase activity in non-target soil organisms

D. Fornasier, N. Mori, P. Tirello, A. Pozzebon, C. Duso, University of Padova / DAFNAE; E. Tescari, Dow AgroSciences Italia srl; R. Bradascio, Dow AgroSciences Italia srl / RD; S. Otto, Italian National Research Council Spray drift of pesticides has a negative impact on aquatic ecosystems and the environment, including damage to non-target organisms. Particularly, the drift of some insecticides can have detrimental effects on beneficial arthropods such as predatory mites. According to the recent EU Directive, the reduction of spray drift is required for a sustainable use of pesticides, yet without reduction of efficacy against pests. In this framework, eight field trials were conducted from 2012 to 2014 in two typical growing areas of Verona district (Northern Italy), four on apple orchards and four on vineyards. The aim of these trials was to evaluate, for two spray drift reduction techniques: 1) the spatial patterns of in-field droplets, 2) the efficacy against predatory mites on apple (Cydia pomonella) and earthworm (Lumbricus terrestris and Dendrobaena octaedra respectively), 3) the side effects on predatory mite populations. Four insecticides, chlorpyrifos, chlorpyrifos-methyl, methoxyfenozide and spinetoram, were applied with three different spraying techniques: high-drift nozzles (Albuz, TVI 80015 green), and high-drift nozzles with an anti-drift adjuvant (rapped oil). Results showed that the two spray drift reduction techniques effectively increased droplets amounts next to spray, reducing potential drift on both apple orchards and vineyards and were generally as effective as standard nozzles without additional side effects on beneficial arthropods. Results suggest that the use of spray drift reduction techniques such as low-drift nozzles and anti-drift adjuvants can be effective in managing key pests and also in decreasing the environmental impact of using insecticides. Full article in: Crop Protection 98 (2017) 283-292, DOI:10.1016/j.cropro.2017.04.010.

TU147 Ariadna spider as a good candidate bioindicator of heavy metal contamination in the Namib Desert

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Human activities are strongly affecting natural ecosystems and native species have been proposed as bioindicators for pollution monitoring and assessment. The present study is the first attempt to use Ariadna spiders as indicators of trace metals contamination in the Central region of the Namib Desert. Mining activity is the biggest contributor to Namibia’s economy in terms of revenue and several trace elements as well as semi-precious gemstones and minerals are main products. Therefore, their released and potential contamination of specific Namib mining areas cannot be excluded. Various spider populations belonging to undescribed Ariadna species are widespread in gravel plains within the Central Namib Desert. Being sit-and-wait predators, Ariadna spiders spend their life in individual tunnels dug in the soil, so resembling the behaviour of ground-dwelling spiders known to be strong metal accumulators in terrestrial ecosystems. In the present study, we collected 60 specimens of three Ariadna populations (20 spiders from each site) in astrual summer 2016, along a N/S and W/E transect at various distances from main mining areas of the Namib Desert. Depth and diameter of entrance burrow and body weight of each spider were recorded. Trace metals analysis were conducted in spider’s whole body as well as in soils samples collected around spider’s burrow. Oxidative stress parameters, CAT, GST and MDA were analysed in soft tissue of spiders and neurotoxicity assessed by measuring cholinesterase activity (ChE). Entrance diameter and depth of burrow seems to be affected by the distance from mining areas. Levels of Zn, Cd, As and Cu resulted higher in Ariadna body compared to levels found in soils around their burrows. On the opposite levels of Pb, V, Cr, Co and Ni were 1 or 2 order of magnitude lower in spiders than in soils. Similar trends in such levels and biological responses as CAT, GST and ChE were observed among sites and based on various distance from the mining area. Such preliminary results support the recognition of Ariadna spider as a good candidate as bioindicator of trace metals contamination in Namib Desert.

TU148 Effect of spray drift reduction techniques on pests and predatory mites in orchards and vineyards

C. Zeiner, Swerea IVF AB / Life Environment; S. Schellenberger, Stockholm University / Department of Applied Environmental Science (ITM); P. Melin, Swerea KIMAB AB; O. Levenstam, University of Borås; A. Hanning, Swerea IVF AB; S. Roos, Swerea IVF AB / Energy and Environment

Microplastic pollution of the marine environment is an environmental issue which is intensely discussed on a global level. Synthetic based textiles contribute to microplastic pollution of the marine environment. Besides littering and the size-shape effects that microplastics have when being exposed to humans and animals they provide an additional vector for chemical pollutants, i.e. possibly providing a new entering mode into organisms of pollutants already existing in the sea. But fibers generated from consumer articles such as textile garments might carry chemical pollutants due to different chemical treatments. This study applies the particle size (PA) and polyester/cotton blend (PES/CO) textiles that were functionalised with durable water repellent (DWR) treatment. The chemical treatment consists of polymers that are based on per- and polyfluoroalkyl substances (PFAS). Question 1: Do we have release of fluorinated fibers from functional textiles? Question 2: What is the amount of fluorinated fibers lost during the washing which can have an impact on the environment? The microscopic investigation identified a new fiber fragment created during the simulation of industrial washing (Gryo wash). Fiber fragments were identified with REM and the EDX analysis of showed fluorine (F) as part of the fiber surface.
composition. The results will be further verified using combustion ion chromatography (CIC) of shedded fibers with and without DWR treatment. These results will then be used to model a scenario simulating for instance Swedish consumption and use of DWR treated outdoor garments and the total exposure of fluorine contaminated fibers to the environment, from washing via waste water treatment to the recipient. This study proved that functional textiles can contribute to the release of microplastic pollution due to the formation of synthetic fiber frayed during washing. The detection of calcium poly(styrene sulphonate) in the fiber such a composition confirmed that these fibers still contained traces of the functional DWR treatment. Released into the environment these fibers might be exposed to long term degradation processes which would finally cause the formation of persistent environmental pollutants. In addition the results of this study suggest that similar mechanism might be relevant for textile containing other functional coatings such as flame retardants, softeners or dyes as well.

TU151
Fate of 14C-labelled Calcium Poly(styrene sulphonate) (CaPSS) Microplastic in wastewater treatment at environmentally relevant concentrations

Microplastics are of high environmental concern due to their widespread occurrence and the fact that so far no sensitive detection systems are available to analyze organic polymers in a complex sludge matrix at such low concentrations. In view of these limitations, the aim of this work was to determine the fate of a model polymer, crosslinked polystyrene sulfonate (PSS), in a simulated WWTP using radiolabeled material. PSS is a polymer which is widely used as an ion exchange resin in various applications. The polymer is insoluble in water and is not degraded in the human body. Calcium loaded PSS (CaPSS) was synthesized in a procedure downscaled from an industrial method with 14C-radiolabelled styrene monomer. This is a key step in the entire project as the radioactivity of the monomer interferes with the polymerization reaction. The resulting 14C-polymer was characterized by comparison with commercial non-labelled CaPSS to prove success. The 14C-radiolabelling enabled detection in sludge matrix as well as the quantification of potential water soluble degradation products and 14CO2 from mineralization. A mass balance was established to identify the most relevant processes for the fate of CaPSS in WWTPs. Due to the high sensitivity of 14C-detection, the test can be performed at realistic/environmental relevant concentrations. As the detection limit of 14C-microplastics in environmental matrices is currently orders of magnitude higher than those detected in the environment, microplastics can serve as an example how future studies on the general topic “microplastics in the environment” can be supported.

TU152
Microplastics in the environment: Evaluating the risks and identifying knowledge gaps
E.E. Burns, University of York / Chemistry; A. Boxall, University of York / Environment Department
The past ten years has seen increasing scientific and public concern over the harmful effects of microplastics (MPs) in the natural environment. In 2010, < 10 scientific publications contained the word ‘microplastic’ while this number had risen to around 170 in 2016. Alongside this, there have been significant policy and regulatory developments around the use and emissions of MPs. We present the results from a systematic review of the published literature to attempt to answer the question “what is the evidence that microplastics adversely impact freshwater and marine systems?” In answering this question, we explore the evidence-base for a number of endpoints detection in sludge matrix as well as the determination of potential water soluble degradation products and 14CO2 from mineralization. A mass balance was established to identify the most relevant processes for the fate of CaPSS in WWTPs. Due to the high sensitivity of 14C-detection, the test can be performed at realistic/environmental relevant concentrations. As the detection limit of 14C-microplastics in environmental matrices is currently orders of magnitude higher than those detected in the environment, microplastics can serve as an example how future studies on the general topic “microplastics in the environment” can be supported.

TU153
A cost-effective methodology for separation of microplastics from freshwater systems
M. R. Rodrigues, Department of Biology & CESAM - University of Aveiro / Department of Biology; F. Gonçalves, University of Aveiro / Department of Biology and CESAM; H. Nogueira, Universidade de Aveiro / Department of Chemistry; J.C. Marques, University of Coimbra / MARE, Dep. of Life Sciences, Coimbra University; A.M. Gonçalves, MARE, Dep. of Life Sciences, Coimbra University/Biologia Department & CESAM, Aveiro University; N. Abrantes, University of Aveiro / CESAM; A. Häusler, GAB Consulting GmbH / Environmental Fate and Modelling

Wastewater is one of the exposure pathways of microplastic into the environment. We have summarized the global coverage of microplastic occurrence studies employing unsuitable analytical confirmation methods which may lead to high error rates and limit data interpretation. In many ecotoxicology studies, results from a systematic review of the published literature to attempt to answer the question “what is the evidence that microplastics adversely impact freshwater and marine systems?” In answering this question, we explore the evidence-base for a number of endpoints detection in sludge matrix as well as the determination of potential water soluble degradation products and 14CO2 from mineralization. A mass balance was established to identify the most relevant processes for the fate of CaPSS in WWTPs. Due to the high sensitivity of 14C-detection, the test can be performed at realistic/environmental relevant concentrations. As the detection limit of 14C-microplastics in environmental matrices is currently orders of magnitude higher than those detected in the environment, microplastics can serve as an example how future studies on the general topic “microplastics in the environment” can be supported.

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We demonstrate that based on current data concentrations of MPs measured in the environment are not high enough to elicit the effects reported from laboratory studies. There is however a mismatch between the size ranges and types of MPs used in laboratory ecotoxicity tests and those detected in the environment. There is an urgent need to address this mismatch by performing better quality and more holistic monitoring studies alongside environmentally relevant effective methods. Only then will we be able to determine whether these materials are having real impacts or not.

TU154
Applicability of remote sensing methods for indirect mapping of microplastic distribution within aquatic ecosystems
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Recently, there have been intensified research efforts to get reliable information about sources, sinks, and transportation pathways of microplastic in aquatic environments. Due to the high spatiotemporal variability of these systems, our knowledge of those aspects is still limited. Earth remote sensing is a key technology within the field of environmental monitoring, providing a unique tool for large area observations of water parameters such as suspended particulate matter, chlorophyll-a or colored dissolved organic matter. Since floating microplastic is probably influenced by the same transport mechanisms as non-motile plankton or motile plankton, we tested the hypothesis of a good detection of microplastic and specific water parameters. In situ water parameters, microplastic (5mm – 250μm), and in situ derived spectral reflectance measurements (ASD FieldSpec) were taken during field campaigns at three different river mouths (Trave and Elbe in Germany, Po in Italy). Microparticles from surface waters were sampled with a manta trawl. Organic material was removed by enzymatic digestion and wet peroxide oxidation, and the remaining microplastics were analyzed down to plastic type with FT-IR as well as SWIR spectroscopy. Initial regression analysis results comparing microplastic with water parameters will be presented, and their suitability as indicators for microplastic abundance is discussed. Furthermore, simultaneously obtained remote sensing data for the river Elbe will be linked with in situ derived microplastic data and water parameters. If significant relationships between microplastic and water parameters exist, remote sensing of water parameters as an indicator for microplastic abundance would provide a cost-effective monitoring tool, able to cover much larger areas than feasible with ship-based point measurement.

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TU155
Coastal accumulation mapping of microplastic particles emitted from the Po River, Italy: Integrating remote sensing, in situ samples and ocean current modelling
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Phytoplankton in inland waters and the open ocean is a long recognized problem for marine life, coral reefs, the fishing industry and shipping transport safety. Microplastics, defined as particles < 5 mm, form a considerable portion of this pollution and have recently received increasing public attention following recent discoveries that not only can these particles be ingested by planktonic animals, but also outnumber natural food items in some ocean areas. Ingested particles can induce negative survival effects as well as serve as introduction vectors for accumulated persistent organic pollutants (POPs) or carcinogenic plastic additives into the base of the food chain, potentially leading to many seafood products consumed by humans. Research has mainly concentrated on marine systems, and while a growing number of studies focus on freshwater lakes, river systems have to date received little attention. In particular, riverine plumes as an important influencing factor for the input and distribution of microplastics into coastal ocean areas remain largely unexplored. Here we present a study of the accumulation of microplastic particles emitted by the Po River along the Adriatic coastline in northern Italy. We posit that river-induced coastal microplastic accumulation can be predicted using a hydrodynamic model, supported by remote sensing data from Landsat and Sentinel-2A. Model accumulation maps were validated against in situ sampling at 9 beaches (analyzed particle size range: 1.5 mm). Hydrodynamic modelling suggests that the amount of discharged particles is only semi-coupled to beaching rates. Object tracking revealed that beaching of emitted particles was strongly mouth dependent and relatively low (less than 25% of all released particles from a given river mouth), primarily occurring within the first five days. The southernmost Po River mouth posed an exception, where more released particles (94%) were found to beach over an extended period of time and along a longer stretch of coastline. Comparison with remote sensing based accumulation maps and validation against in situ beach sampling are discussed. The presented methodology lays the groundwork for developing an operational monitoring system to assess plastic micropollution being emitted by a major river and its distribution along adjacent coastlines as well as into the open ocean.

TU156
Cause and effect of the plastic industry in South Africa as a developing country
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In 2017, the South African plastic industry has grown with 1.9%, compared with 2016 (Plastic value of 6.3bn R). The plastic industry in South Africa as a developing country has set an ideal of no plastics to landfills by 2030, and plans are being set in place to up efforts. Much effort is spent corporately to reduce South Africa’s ‘plastic footprint’, but efforts in terms of microplastics are trailing. We collected and clean samples from a given river mouth), primarily occurring within the first five days. The southernmost Po River mouth posed an exception, where more released particles (94%) were found to beach over an extended period of time and along a longer stretch of coastline. Comparison with remote sensing based accumulation maps and validation against in situ beach sampling are discussed. The presented methodology lays the groundwork for developing an operational monitoring system to assess plastic micropollution being emitted by a major river and its distribution along adjacent coastlines as well as into the open ocean.

TU157
Understanding the distribution and fate of microplastics in a tertiary sewage treatment plant in the UK
R.M. Blair, S. Waldron, University of Glasgow; C. Gauchotte-Lindsay, University of Glasgow / Infrastructure and Environment; V. Phoenix, University of Strathclyde / Civil and Environmental Engineering; Microplastics (MPs; < 0.5 mm) are classified as contaminants of emerging concern but currently are not regulated by water quality standards. Microplastics are highly diverse and their distribution in the environment is highly variable in space and time, making their quantification and risk assessment difficult. Further, their monitoring and regulation are hindered by limited empirical data, particularly of freshwater and wastewater systems as important pathways of land-based contaminants to oceans. Here, a study was conducted in a tertiary sewage treatment plant in the UK (Glasgow, Scotland) to assess the presence of MPs in the system and the effect of treatment stage in removing these contaminants before discharge into recipient freshwater. The MPs were extracted from 5 L mixed liquor samples taken at five treatment stages, using H₂O₂ digestion and vacuum filtration through 1.2 μm GF filter. Characterisation and quantification of MPs was carried out by light microscopy followed by detailed chemical analysis of representative subsamples via SEM-EDS and FTIR-ATR. Microplastics were present in wastewater samples collected at all treatment stages, and abundances generally decreased from inflow to outflow. Further, high variability in the concentrations was evident across sampling dates. Chemical characterisation by SEM-EDS revealed that 94% of analysed pieces were C-based materials, but only 25% were confirmed plastics based on FTIR-ATR results. In conclusion, the tertiary treatment process evaluated here efficiently removes MPs entering the system but small quantities may still be discharged into the environment. Further, visual characterisation with light microscopy may result in overestimation of MPs due to the misidentification of cellulose and other natural fibres as plastics. Therefore, analysis of chemical composition through a combination of detailed analytical techniques is crucial for improved accuracy of results. This study contributes to understanding what methods are needed to extract and identify MPs from environmental samples, and the spatio-temporal data generated provide understanding of what needs to be monitored and where controls should be implemented.

TU158
Weathering-induced changes in the effects of microplastic particles and their leachates
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Studies on the potential effects of microplastic (MP) particles in the aquatic environment are numerous. However, many laboratory studies apply spherical, pristine particles, which may be of limited relevance given UV light irradiation, mechanical stress, salinity, biofilm growth and other factors that can influence the weathering of the particles. To the authors’ knowledge, there are no studies that have investigated the weathering of MP particles in a marine environment. The Oceans-funded project WEATHER-MIC is to assess the impacts that weathering has on the transport, fate and effects of MP particles and their leachates. We summarize recent results on potential effects. (1.) Impact of MP particles on organisms: We have exposed copepods, daphnia and algae to different fractions of virgin and weathered MP as well as particle-free leachates under controlled conditions. From the observation of apical endpoints in the acute toxicity assays, concentration-response relationships for the different fractions can be deduced. A critical evaluation of the suitability of the applied test protocols for the assessment of adverse effects of MP will be presented. (2.) Influence of ageing plastic and leachates on biofilm structure and function: Natural biofilms (containing bacteria, algae and fungi, embedded in extracellular polymeric substances) grown on various microcosms on different types of aged and pristine polymeric substrates have been studied to observe the influence of weathering on the attachment and succession of biofilms. Sum parameters (biomass, pigment profiles, photosynthesis) and sequencing data were studied. (3.) Mixture effects of leachates from the most common polymers: Cell-based bioassays have been applied to study mixture effects of additives and degradation products of the polymers. This study has been conducted during weathering of plastic material in artificial seawater in agitated UV chambers. The chemicals in the seawater leachates were enriched by solid-phase extraction or chemicals were directly extracted from pristine particles by ultrasonic-assisted solvent extraction. The concentrated leachates and solvent extracts were then dosed into cell-based bioassays, covering i) cytotoxicity; ii) acute effects of metabolic toxicity, e.g. via binding to intracellular carbon receptor; iii) specific, receptor-mediated effects such as endocrinology; and iv) adaptive stress responses such as oxidative stress. The results may help to understand effects caused by additives and parent compounds opposed to the degradation products liberated from the UV-weathered plastic.

TU159
Occurrence and characteristics of fine microplastics in sewage water, domestic water, sewage treatment water and river water by coagulation and FT-IR microscopy method
Y. Kameda, Chiba Institute of Technology / Creative Engineering; N. Yamada, Chiba Institute of Technology; The occurrence of Microplastics (MPs) is of great concern in aquatic environment, especially ocean. Many current studies evaluate MPs ranged from 100 μm to 5 mm. However, MPs used in personal care products and other industrial processes are reported to be smaller according to previous reports. Because MPs are very...
important substances to current economic activities, new materials for MPs will be needed such as cellulose. Though alternative of the materials is mainly conducted in personal care products, effect of the volunteer actions and various regulations on decrease of MPs in aquatic environments has not been evaluated. It is necessary to reveal their sources such as sewage water, sewage treatment water, wastes in aquatic environments and so on. This research shows occurrence and characteristics of MPs in sewage water, sewage treatment water, runoff river water by coagulation and FT-IR microscopy method developed in our laboratory. MPs in the various contaminated water were collected by a plankton net whose mesh size is 10 µm. The collected particles were separated by a density separation method. After that, MPs in the collected particles were separated by coagulation process. Finally, the MPs were passed through a membrane and were identified by standard adding microplastics MPs by FT-IR microscopy. MPs are characterized by materials, size, color and multi-regression analysis by FT-IR spectrum data. Based on these data, contribution of MPs from personal care products to total MPs concentration will be discussed as well as estimation of sources of MPs in various water samples.

TU160
Detection of micro-paint particles and microplastic in harbour soil samples using FPA-µFTIR-Imaging-FTIR
A. Vianello, Aalborg University / Civil Engineering Department - Section of Water and Environment; M. Simon, N. van Alst, F. Liu, Aalborg University / Civil Engineering Department; K.B. Olesen, Aalborg University / Department of Civil Engineering (25%), ally coated Aalborg University / Civil Engineering Department

Microplastic (MP) pollution is nowadays recognized as a global concern affecting both marine, freshwater and also the terrestrial environment. Beside microplastic pollution, also micro-paint particles (MPP), originated by the degradation and abrasion of painted surfaces (buildings, ship-paint materials) have been reported in some recent studies. As many paints also contain organic compounds and heavy metals mixed as biocides aimed to inhibit the growth of biota on painted surfaces, these particles could be even more harmful to the environment and wildlife than microplastics. Here we present some results obtained from the analysis of soil samples collected in a recreational boat facility in the North of Denmark. A “Microplastic-based” approach has been used, extracting the particles from the soil matrix using air-assisted density separation followed by FPA-µFTIR-Imaging analysis. This approach allows to identify and quantify microplastics and micro-paint particles down to 10–20 µm in size. Surficial soil samples were collected along three transects located in different areas of the shipyard. The samples, previously sieved (5000–500 µm and 500–100 µm) were submitted to flotation using ZnCl₂ followed by sample cleanup using enzymes and H₂O₂ oxidation to remove organic matter. The analysis was carried out using FPA-µFTIR-Imaging spectroscopy and the data were processed with a dedicated software (MPHunter) developed at Aalborg University. The first results highlighted a high micro-paint and microplastic particles contamination. The total MP and MPP concentration were 222,500 particles Kg⁻¹, while the estimated mass was 17.1 mg Kg⁻¹. The most abundant polymers/paints detected were polyestere (30%), acryl coating (20%) and polyethylene (17%). The particle size distribution showed the most abundant size ranges were between 20–40 µm and 40–80 µm. The high MP and MPP concentration measured in the sample highlights the harbor areas are potential hotspots for the accumulation and further spreading of MP and MPP in the terrestrial and aquatic environment. Microplastic and micro-paint particles were successfully extracted and detected in a recreational harbor using the art analytical approach including multi-step sample preparation and FPA-µFTIR-Imaging analysis, the most suitable spectroscopic technique for an unbiased MP detection and quantification.

TU161
Runoff of microplastics from agricultural soil: a study in a semi-arid area
R. Harvey, NIVA - Norwegian Institute for Water Research; T.C. Schell, IMDEA Water Institute / Ecotoxicology; R. Hurley, Norwegian Institute for water research; A. Rico, IMDEA Water Institute / Aquatic Ecotoxicology; M. Vighi, IMDEA Water Institute / Earth and Environmental Sciences

Wastewater is considered to be one of the major sources of microplastics (MPs) entering surface waters. Although a high retention potential of wastewater treatment plants (WWTPs) for MPs in raw wastewater has been reported, the composition using terms such as spectroscopy. Finally, the more erratic pristine characteristics of the watershed (total population, characteristics and location of WWTPs, agricultural land use patterns including sludge application, etc.) will be integrated into the study’s findings to deduce their importance for MP contamination. The preliminary results of the distribution and characterization of MP in different matrices are presented.

TU162
Microplastics in wastewater and freshwaters: a case study in the Henares river watershed (Central Spain)
T.C. Schell, IMDEA Water Institute / Ecotoxicology; R. Hurley, Norwegian Institute for water research; A. Rico, IMDEA Water Institute / Aquatic Ecotoxicology; M. Vighi, IMDEA Water Institute / Earth and Environmental Sciences

Microplastics occurrence and composition in drinking water from a Norwegian urban area
a. gomiero, International Research Institute of Stavanger / Environment; G. Skogerbo, IVAR; K. Øysæd, A. Vatland Kruvel, International Research Institute of Stavanger

Microplastics as a potential health and environmental problem has gained increasing attention recently. Microplastic is defined as plastic pieces smaller than 5 mm in diameter, and the sources of microplastic are many. State of the art literature reports that microplastics are ubiquitous worldwide. While several authors report fragmentation of different plastic types and microplastics in the aquatic, freshwater, marine and terrestrial compartments of marine, freshwater and terrestrial ecosystems; others point out the accumulation of micro- and nanometric sized plastic particles thorough the marine and terrestrial food webs posing the risk of marine and terrestrial life and ultimately the human health. Despite of these studies point out the occurrence of micro plastics in freshwater systems including surface and groundwater basins, very little in know about occurrence of microplastics in the drinking water and their implications on human health. According to the WHO men should consume 3 L and women should consume 2.2 L of beverage per day. Most of these beverages consist of tap water, or drinks derived from tap water (such as coffee, tea, or reconstituted juice). The risk of plastic uptake from drinking water is currently unpredictable and furthermore, these plastic particles are in addition to plastics potentially consumed in other sources, such as sea salt, beer, and food and seafood. The research tasks of the present work were: Map published and available literature, develop and optimize a standardized fast, sensitive protocol for sampling and quantification of nano/microparticles in drinking water and finally. Analyze and possibly
detect microplastic particles in drinking water supply systems with special focus on different polymeric composition and size fractions. Study area was the Rogaland area (Norway) populated by approx 110,000 inhabitants. Samples of drinking water were collected every two week for ten months contemporary from the supply water system collection point as well as in different sites of the urban area. A fast and sensitive method based on a GCMs-pyrolysis was developed. Polyethylene, Polypropylene and Polyvinyl chloride were the most recurrent polymers. Levels ranged from 0.02 to 16 ng/L. Time and space related trend are presented.

TU164 Macro and Micro(plastics) in the Environment of Some French rivers
V. Verney, CNRS- ICCF/Photochimie-CVP; G. BISSAGOU KOUMBA, UNIV Nantes, F. Delor Jestin, SIGE-ICCF; Z. Dominink, H. Askarian; CNRS-ICCF; J. Peiry, E. Roussel, O. Voldoire, CNRS-Geolab; A. Schaal, L. Durantou, Observatoire du Microplastique; M. Liboiron, Memorial University of Newfoundland

It is now known that the vast majority of microplastics found in the seas and oceans originate from lands. In such a process fresh water environment (rivers and riverbanks) play a major role. It is therefore necessary to imagine the scenario that a used plastic, becoming a waste after use, will be found in the environment if it has escaped to a waste treatment stream. Its stay in the environment can persist for a very long time and this waste will then be exposed to a set of environmental constraints (UV, rain, wind, mechanical erosion,…) which will continue and amplify its degradation, leading to its fragmentation. The work we have undertaken consists of: 1. Mapping the presence, the mobility and the chemical composition over time of macroplastics present on the banks of an experimental site of the Allier River, and linking it to the density of the vegetal areas. 2. Set up on site a controlled pollution to follow its fate along the time. 3. Analyze the composition of microplastics extracted from the sediments, especially at the entrance of the abandoned channel, where it may exist some vortices of flow. These first three points are the topic of the Plasticscages project supported by the CNRS[1, 2]. 4. Collect and analyze the composition of microplastics in the surface waters of different French rivers (Allier, Charente, Loire, Touvre, etc.). To do this, we rely on citizen science operations, in particular thanks to the contribution of the babylleg sampling net [3,4], which makes it possible to multiply samples and analyses. 1 Occurrence of plastic litter in the Allier river in France. Vincent Verney, Gaëlle Bissagou Koumba, Alexandre Garreau, Florence Delor, Olivier Voldoire, Jean-Luc Peiry; To be published 2- https://www.researchgate.net/project/PLASTICSCAGES 3- Compromise agency, the case of babyllegs, Max Liboiron, Engaging Science, Technology and Society (2017), 499-527 4- http://lapagaisesauvage.org/laboratoirerecyclen/

TU165 Spatial and temporal trends of microplastics in an urbanized Canadian river
M.S. Ross, T. Bujaczek, S. Koter, MacEwan University / Department of Physical Sciences; D. Locky, MacEwan University / Department of Biological Sciences

Microplastics are ubiquitous contaminants in the marine environment, but quantification is limited in both freshwater systems and urban environments. While it is now recognized that microplastics are dispersed along the full continuum of aquatic environments, and that urban centres are major sources, the extent to which microplastics are transported to freshwater systems remains underexplored. This study examines the occurrence, composition, and potential sources of microplastic contamination in the North Saskatchewan River, an urbanized river flowing through the city centre of Edmonton, Alberta, the fifth largest city in Canada. Surface water samples were collected monthly during the summer of 2017 using plankton nets with 53µm mesh. Samples were collected from seven sites throughout the watershed (upstream of the urbanized city and potential point sources i.e., a wastewater treatment plant, WWTP). Microplastics were found in all samples, and at some sites concentrations exceeded those reported in other urbanized rivers. Various coloured fragments, films, beads, and fibers were identified, with the majority of microplastic contamination being in the form of fragments. Both the total concentrations and the proportion of each type of plastic varied with distance downstream and size class, suggesting changes in the river flows through the city. No differences in either concentration or composition were found between sites upstream and downstream of the WWTP, suggesting a lack of significant input to the river. Moving forward, the chemical composition of suspected microplastics will be investigated using Raman microspectrophotometry.

TU166 Models for Data Synthesis, Sampling Design and Scenario Assessment: Some using the INCA-MP model of microplastic fate and transport in soils and surface waters
M. Futter, Swedish University of Agricultural Science / Aquatic Sciences and Assessment; J. Crossman, University of Windsor; J. Ledesma, V. Russo, E. Lennegård, SLU Swedish University of Agricultural Sciences / Aquatic Sciences and Assessment; L. Nizzetto, NIVA

Quantification and classification of microplastics in soils, sludge and surface waters is both time consuming and expensive. Ideally, measurement campaigns can be focussed on areas that are likely to provide the greatest returns on effort yet this is often difficult to accomplish. Here we show how the Integrated Catchments model for Micro Plastics, the first published model of microplastic terrestrial fate, riverine transport and contaminant co-transport can be used to synthesize available data, identify knowledge gaps, plan monitoring, and perform risk assessments. Synthesizing available data involves collation of microplastic and proxy data. We show how proxy information, including timing and integrated exposure based measures of fibres and high frequency water quality monitoring can constrain estimates of microplastic mobility in terrestrial and freshwater environments. Through the application of uncertainty analysis in INCA-MP, it is possible to identify the most sensitive pools and processes when making predictions of microplastic fate and transport. Furthermore, knowledge gaps related to these pools and processes can then be targeted for more intensive field sampling campaigns. As the INCA-MP includes sophisticated routines for tracking the environmental fate of current and emerging micropollutants, the model can be used for risk assessment of co-transported contaminants. Such information is needed both for more targeted sampling.
campaigns and for communicating risks associated with microplastics in terrestrial and freshwater environments. We illustrate these concepts using data from Swedish and Canadian catchments. Our results show the importance of autumn storms and spring snowmelt for plastic mobilisation to surface waters and highlight the potential knowledge gaps associated with targeted sampling of riverine sediments, constructed wetlands and waste treatment facilities.

TU169 Occurrence and concentration of microplastics in an urban river C. Campanale, C. Massarelli, G. Bagnuolo, Italian National Research Council; V. Uricchio, Italian National Research Council / Water Research Institute

The term ‘microplastics’ was first used in 2004 to describe very small fragments of plastic (up to 50 μm) in the water column and in sediments. In 2012, 3.52 to 13.43 μm showing significantly higher abundances during February than April campaign (Mann–Whitney U Test = 18.00; p-value = 0.028). A total of five polymer have been characterized: PE, PP, PS, PVC and TD-PUR. All samples contained at least three polymer types: PE, PP and PS. PE accounted for 77% of the total particles identified, followed by PS (12%), PP (10%), PVC (0.9%) and PU (0.4%).

TU170 Removal of 10-500 μm microplastics from wastewater effluent by disc filter M. Simon, Aalborg University; N. van Alst, Aalborg University / Civil Engineering Department; K.B. Olesen, Aalborg University / Department of Civil Engineering; F. Liu, J. Vollertsen, Aalborg University / Civil Engineering Department

In this study the efficiency of a disc filter to remove microplastic concentrations from wastewater effluent was evaluated. The size range of particles addressed was 10-500 μm and the identification technology was micro-FTIR imaging spectroscopy applying a focal plane array (FPA). Effluent wastewater was collected at the wastewater treatment facility at Grindsted, Denmark operated by Billund Spildevand A/S. The treated wastewater was sampled before and after the disc filter by means of a single 10-m² stainless steel continuous flow large-scale water sampling device. The filtered volume of effluent wastewater before the filter was 200 L and 1.6 m³ after the filter. The residue collected on the filters containing a mixture of organic matter, inorganic particles and microplastics was subjected to a purification procedure including enzymatic digestion, chemical oxidation and filtration in order to eliminate the sample matrix and extract the microplastics. Non-degradable particles were stored in ethanol, and a fraction of the ethanol particle suspension transferred to a transmission window to quantify particles by infrared imaging technique. The entire window was scanned to create a mosaic with 3.3 μm pixel resolution on the FPA. The spectra of all particles in a scan were analyzed to quantify their chemical composition and to determine whether they were of plastic, and if so, of which plastic material. The spectral analysis was carried out with a semi-automated IR spectra analyzer software developed at Aalborg University, Denmark. The size and shape of plastic particles were recorded and their mass was estimated. Preliminary data shows that the removal efficiency of the disc filter was 96% in terms of both mass and particle number. The material composition of plastic in the sample before the filter (polystyrene, polyethylene and acrylates) was somewhat dissimilar to the composition in the effluent sampled after the filter (polystyrene and polystyrene).

TU171 PlasticBudget - Project on the environmental assessment of microplastic emissions N. Thomann, Fraunhofer Institute for Environmental, Safety, and Energy Technology UMSICHT

Shortly after the introduction of many types of plastics (e.g. polystyrene, polyethylene terephthalate, polypropylene) in the 30s, 40s and 50s, first traces of plastics in the environment have been detected. Packaging, pellets and parts of a kitchen sponge were found in the stomachs of seabirds; Whales and seals were caught in polypropylene cords. By weathering and fragmenting larger plastic objects (macrophastics) into smaller pieces (microplastics), plastic waste in the environment seems to be gradually disappearing. However, as recent research shows, microplastic is found in freshwater, on beaches and in open water, in the deep sea and in the Antarctic ice. It is taken up by organisms and processes in the food chain. The long discussion of the methods suggested the plastic waste will accompany, if not outlast, mankind for a long time to come. Although the number of publications on microplastics has risen in the last two decades and the topic has entered the social discourse, there are still many research gaps on sources, pathways, amounts, sinks, accumulation spaces, adsorption and absorption of pollutants as well as damaging effects on organisms and humans. The project PlasticBudget is aiming to close some of the above-mentioned research gaps. Taking into account the relevance that plastic litter has gained in recent years in the environmental discussion, the assessment of the environmental impact of those emissions is needed. Macro- or microplastics’ emissions have an impact on ecotoxicity (for example, when birds or fish confuse plastic with food) and human toxicity (for example by eating food, which contains microplastics). The resulting environmental impacts could be, for example, the extensively documented dying of marine organisms by microplastics, the danger of massive aggregation of tiny plastic particles in the food chain, or even the negative aesthetic impact associated with plastic in the environment. Corresponding midpoint and endpoint indicators and associated characterization methods, as well as standardization to a reference value (e.g. by the production volume of the specific plastic), are therefore developed in the PlastikBudget-project and discussed in expert dialogues as well as workshops.

TU172 How do we know that microplastics are different from natural particles in their effects on biota? Z. Gerdes, M. Ogonowski, E. Gorokhova, Stockholm University / Department of Environmental Science and Analytical Chemistry (ACES)

Microplastics (MP) have been identified as a potential environmental hazard, which has motivated a wide range of effect-studies, testing different combinations of polymer materials and shapes. However, risk assessment of MP exposure, in the lower size range < 100μm, is today hampered by both the lack of data regarding their presence in the environment and the inadequate experimental design of many effect-studies. A crucial issue in designing such studies is to include control particles that are ubiquitously present in the environment and represent a background variability with regard to suspended solids. Such controls, allow effects caused by the particle per se to be separated from that caused by specific chemical contaminants and are hence a factor that needs to be considered if the environmental fate of pollutants is examined. Some contaminants, among those also micropollutants like pharmaceuticals, can be charged under certain pH conditions. While it is known for a wide variety of natural particles that charged compounds sorb only little or not at all, sorption interactions between microplastics and charged compounds have not been analysed so far. Thus, the aim of this study was to clarify the sorption behaviour of dissociating compounds to microplastic particles. We measured the equilibrium partitioning between 19 typical wastewater contaminants (pharmaceuticals, personal care products, pesticides) and microplastics at three different pH conditions (4.7, 7.0 and 9.0). The investigated compounds showed a wide variety in their physico-chemical properties, e.g. a log Kow range between 0.1 and 5.8 and pK_a values from 1.6 to 13.9. We performed batch experiments with fourteen ionicizable and five non-ionicizable substances. In all experiments equilibrium was reached after two days. Measured log Kow for the neutral species ranged from 0.75 to 4.00. The uptake of contaminants varied according to their hydrophobicity. Sorption of ionicizable substances is strongly influenced by the pH while non-ionicizable substances showed a partitioning independent of pH. For sorption into polyethylene, the amount of accumulated pollutants is principally dominated by the neutral fraction, while the charged
species did not contribute. Thus, with increasing pH sorption of acids decreased while the sorption of bases increased. Whereas electrostatic interactions between charged species and polyethylene could not be detected, this might be different for other polymers, such as polyethylene and polyamide.

TU174 Influence of microplastics on transport of organic contaminants in soil
T. Hüffer, S. Slawek, T. Hofmann, University of Vienna / Department of Environmental Geosciences
The worldwide production and usage of mainly disposable plastic has increased from 1.7 million tons in 1950 to 299 million tons in 2013 [1]. Consequently, plastic wastes are deposited in the environment and persist due to long durability and limited recovery [2]. Polyethylene is one of the mass-manufactured polymers that is found in the terrestrial environment, used in many different sectors, including agricultural mulches, composites and packaging material [3]. To date, microplastics have been mainly studied in marine and freshwater systems, while there is hardly any data on microplastic occurrence, fate, and effect in terrestrial environments [2]. Low-density polyethylene (LDPE) foils, that may become brittle due to isolation, are used in large amounts on agricultural areas to protect crops, suppress weeds, regulate the temperature and retain irrigation water in the soils [1]. In soil microplastics may affect the transport of hydrophobic organic pollutants and pesticides, as they can be preferentially sorbed by polymer particles with large surface to volume ratio compared to sorption by natural sorbents [1, 4]. The strength of sorption as well as the relevant molecular interactions depend on the properties of the polymer, for instance because they can incorporate [5]. Therefore, this study investigates the influence of microplastics on the transport of organic contaminants in a soil under varying aqueous conditions. [1] K. Dus, A. Coors, Environ. Sci. Eur. 2016, 28, 2. [2] M. Rillig, Environ. Sci. Technol. 2012, 46, 6453. [3] M. Beg, S. Kormin, M. Bijariim, H. Zaman, Adv. Polymer Technol. 2015, 35, 2152. [4] A. Bakir, S. Rowland, R. Thompson. Mar. Pollut. Bullet. 2012, 64, 7282. [5] T. Hüffer, T. Hofmann, Environ. Pollut. 2016, 214, 194.

TU175 Influence of polystyrene microplastics in combination with organic pesticides on the giant rams-horn snail Marisa cornuarietis: behavioral and biochemical responses
S. Krais, University of Tubingen / Animal Physiological Ecology; H. Schmiegel, Tübingen University / Animal Physiological Ecology; E.E. May, University of Tuebingen / Animal Physiological Ecology; A.S. Ruhl, TU Berlin / Department of Water Quality Control; H. Köhler, University of Tubingen / Animal Physiological Ecology; R. Triebkorn, University of Tuebingen / Animal Physiological Ecology. D. Prada, E. Gorokhova, Stockholm University / Department of Environmental Science and Analysis; D. Prada, E. Gorokhova, Stockholm University / Department of Environmental Science and Analysis. Microplastics are of particular interest in aquatic systems like pesticides or pharmaceuticals, transport them into food chains and modulate their toxicities. In addition, they can mechanically affect exposed organisms. Whereas in the past, most of the studies on microplastics have focused on the marine environment, there is still little knowledge about the occurrence and impacts of microplastics in freshwater ecosystems. The aim of this study is to examine possible influences of polystyrene particles in combination with different organic pesticides on the behavior and biochemical responses of the giant rams-horn snail (Marisa cornuarietis). Snails were exposed to 10,000 polystyrene particles per liter (cogenetically milled, < 100 μm) in combination with different concentrations of the pesticides cypermethrin, methiocarb and thiacloprid. In order to quantify the observed behavioral responses, five categories of behavior were defined, which are “crawling”, “attached to the wall”, “attached above the water surface”, “inactive on the ground” and “retracted with closed operculum”. All snails were individually categorized twice a day for nine days. The results make evident that snails exposed to cypermethrin significantly changed their behavior between the first (day 1-4) and second (day 5-9) observation period, independent of the test concentration. In the first period, they were often categorized as “attached to the wall” or “attached above the water surface”, whereas in the second observation period, these snails were mainly classified as “inactive” or “retracted”. As biochemical endpoints we study oxidative stress (lipid hydroperoxides, superoxide dismutase), proteotoxicity (Hsp70 level) and neurotoxicity (inhibition of acetylcholinesterase). The analyses, however, are still in progress. The study is part of the joint research project MiWa (“microplastics in the water cycle”) funded by the German Federal Ministry of Education and Research (support code: 02WS1645).
Analytical Chemistry (ACES)
Conventional, oil-based polymers are considered as the major source of microplastic pollution, whereas biodegradable polymers (bioplastics) have not attracted much attention as sources of microplastics. However, given that production of bioplastics is increasing and that biodegradation is slow under ambient conditions, it is likely that they enter the aquatic environment in the same way as other biodegradable polymers. Therefore, it is important to understand potential environmental impacts of both polymer types. We compared effects of exposure to polylactic acid (PLA; biopolymer) and polystyrene (PS; oil-based polymer) on primary life history traits in the crustacean *Daphnia magna*, a standard model species in ecotoxicology. To exclude particle effects caused by food dilution and thus identify microplastic-specific effects, kaolin clay was used as a reference treatment. In total, four treatments were included: PLA, PS, clay (reference), and control (food only). The exposure was conducted over 21 d using a plankton wheel to keep test particles and algae in suspension for comparable exposure concentrations. In the PS treatment, we observed high mortality, decreased feeding rate and reproductive output compared to all other treatments. These effects were not caused by toxic monomers of styrene or additives leaching out of the polymer, which was demonstrated in a follow-up test with the PS leachates. By contrast, no significant effects were found in the daphnids exposed to PLA compared to the reference treatment. Thus, a significantly higher toxicity of the conventional polymer was observed, whereas effects of the biodegradable microplastics were similar to those caused by the ubiquitously occurring clay particles. More studies are needed to identify the mechanisms of PS toxicity and to confirm the observed ecotoxicological findings, especially due to the low toxicological potential of the polymer type used during testing.

To evaluate toxicity of these materials, it is essential to include a reference treatment as a benchmark.

TU179 Effects of polystyrene microplastics in different life stages of brown trout (Salmo trutta f. fario)
H. Schmieg, Tübingen University / Animal Physiological Ecology; S. Krais, University of Tubingen / Animal Physiological Ecology; F. Rezbach, University of Tubingen / Animal Physiological Ecology; A.S. Ruhl, TU Berlin / Department of Water Quality Control; H. Köhler, University of Tubingen / Animal Physiological Ecology; R. Trubenkov, University of Tubingen / Animal Physiological Ecology. The widespread use of plastic products in our daily life has led to a constant increase in the production of synthetic polymers. In consequence and also resulting from the longevity of plastics, high amounts of plastic debris can be found worldwide in aquatic and terrestrial environments. In general, plastic items smaller than 5 millimeters are defined as microplastics. Primary microplastics are produced for different purposes and are, for example, contained in many cosmetic products. Abrasion and fragmentation of larger plastic items lead to the formation of secondary microplastics. Up to now, most studies investigating effects of microplastics in organisms concentrate on marine ecosystems, whereas knowledge on effects of microplastics in freshwater organisms is still scarce. The aim of our study is to investigate effects of polystyrene microplastics (cylindrically milled granules, fractionated to < 0.8 μm, up to 100,000 particles/L) in different life stages of brown trout (Salmo trutta f. fario). For that purpose, we conducted a fish early life stage test (FELST) according to OECD 212. The endpoints of interest were heart rate, hatching success and mortality. After consumption of the yolk sac by the fish larvae, we additionally investigated the level of oxidative stress by means of the fluorescent dye heisenburg (nAcute) bound to the fish larvae. We examined effects of polystyrene particles (< 50 μm, 10,000 particles/L) alone and in combination with organic pollutants (pharmaceutical, pesticide) in different life stages of brown trout. In this experiment, the mortality rate, biometric parameters, the level of oxidative stress, the induction of the 70 kD stress protein (Hsp70) and the inhibition of acetylschioninesterase were under investigation. Furthermore, we examined histopathological effects in gills and in guts of the trout. First results showed no effect of microplastics on mortality and biometric values of either larval or juvenile brown trout. Further analyses are still in progress. The present study is part of the joint research project “MiWa” (microplastics in freshwater systems) funded by the German Federal Ministry of Education and Research (support code: 02WR1S1378).

TU180 Daphnids in distress? Acute and chronic effects of primary and secondary microplastics on three species of Cladocerans
D. pulex in distress? Acut T. Örlof, U. P. Steinhart, M. W. Schmieg, J. M. Schmieg. University of Tübingen / Animal Physiological Ecology; F. Rezbach, University of Tübingen / Animal Physiological Ecology; S. Krais, University of Tübingen / Animal Physiological Ecology; H. Köhler, University of Tübingen / Animal Physiological Ecology; R. Trubenkov, University of Tübingen / Animal Physiological Ecology. The widespread use of plastic products in our daily life has led to a constant increase in the production of synthetic polymers. In consequence and also resulting from the longevity of plastics, high amounts of plastic debris can be found worldwide in aquatic and terrestrial environments. In general, plastic items smaller than 5 millimeters are defined as microplastics. Primary microplastics are produced for different purposes and are, for example, contained in many cosmetic products. Abrasion and fragmentation of larger plastic items lead to the formation of secondary microplastics. Up to now, most studies investigating effects of microplastics in organisms concentrate on marine ecosystems, whereas knowledge on effects of microplastics in freshwater organisms is still scarce. The aim of our study is to investigate effects of polystyrene microplastics (cylindrically milled granules, fractionated to < 0.8 μm, up to 100,000 particles/L) in different life stages of brown trout (Salmo trutta f. fario). For that purpose, we conducted a fish early life stage test (FELST) according to OECD 212. The endpoints of interest were heart rate, hatching success and mortality. After consumption of the yolk sac by the fish larvae, we additionally investigated the level of oxidative stress by means of the fluorescent dye heisenburg (nAcute) bound to the fish larvae. We examined effects of polystyrene particles (< 50 μm, 10,000 particles/L) alone and in combination with organic pollutants (pharmaceutical, pesticide) in different life stages of brown trout. In this experiment, the mortality rate, biometric parameters, the level of oxidative stress, the induction of the 70 kD stress protein (Hsp70) and the inhibition of acetylschioninesterase were under investigation. Furthermore, we examined histopathological effects in gills and in guts of the trout. First results showed no effect of microplastics on mortality and biometric values of either larval or juvenile brown trout. Further analyses are still in progress. The present study is part of the joint research project “MiWa” (microplastics in freshwater systems) funded by the German Federal Ministry of Education and Research (support code: 02WR1S1378).

TU181 Evaluation of chronic toxicity of polystyrene microplastics on freshwater mussels
S. Magni, University of Milan / Department of Biosciences; F. Gagne, Environment and Climate Change Canada; C. Delta Torre, State University of Milano / Biosciences; C. Andrè, J. Auclair, H. Hanana, Environment and Climate Change Canada / Aquatic Contaminants Research Division; F. Bonasoro, University of Milano / Department of Environmental Science and Policy; A. Binelli, University of Milano / Department of Environmental Science and Policy; A. Binelli, University of Milano / Department of Environmental Science and Policy; A. Binelli, University of Milano / Department of Environmental Science and Policy; A. Binelli, University of Milano / Department of Environmental Science and Policy; A. Binelli, University of Milano / Department of Environmental Science and Policy. The annual global plastic production follows a positive trend and plastic pollution represents an emerging worldwide issue. In particular, microplastics (MPs), plastic fragment smaller than 5 mm, are potentially dangerous for aquatic communities because their ability to be accumulating in the tissues of biota. MPs can reach the aquatic environment through the Wastewater Treatment Plants (WWTPs) or affects due to the degradation of microplastics. Considering that few studies, especially in freshwater environment, have been conducted about the adverse effects of MPs, the aim of our study is the evaluation of chronic toxicity of these contaminants on the freshwater mussel *Dreissena polymorpha* using a multi-biomarker approach. As MP standards we choose two different beads of polystyrene, one of the most common MP classes detected in the environment, with a size of 1 and 10 μm. On the basis of the daily great release of MPs from WWTPs, we tested the following mixtures (MIXs) of polystyrene MPs: MIX1, which contained 2 millions/L of 10 μm MPs and 2 millions/L of 1 μm MPs, and MIX2, which contained 500,000/L of 10 μm MPs and 500,000/L of 1 μm MPs. Therefore, mussels were exposed for 7 days in static conditions to the MIXs and to related controls; every 3 days we collected from each tank the mussels to assess both chronic toxicity and uptake of polystyrene MPs. We evaluated the adverse effects by monitoring end-points of cellular stress, as the activity of antioxidant and detoxifying enzymes, oxidative damage, cytogenotoxicity and neurotoxicity (analyses in progress). To evaluate the uptake of polystyrene MPs in the exposed mussels, exploiting the reflexion of MPs, we collected hemolymph and then fixed them for 21 d using a cryostat sectioning. We then observed the samples using the confocal microscopy. Despite we found both sizes of polystyrene MPs in the hemolymph and soft tissues of mussels, we did not obtain significant increase of tested biomarkers compared to control, excepted for a significant increase of CAT activity at the end of exposure for mussels exposed to MIX 2. The lack of adverse effects induced by tested MPs could suggest that the toxicity of MPs could be related to some degradation processes of MPs towards chemicals, or involved in metabolic pathways not detectable by our biomarkers. In addition, prolonging the exposure time the MP toxicity could be increased.

TU182 Polystyrene microplastic effects on the lipid peroxidation and antioxidant capacity in non- and temperature-stressed individuals of *Dreissena polymorpha*
A. Weber, N. Jeckel, C. Weil, S. Umbach, Goethe University Frankfurt am Main / Aquatic Ecotoxicology; N. Brennholt, German Federal Institute of Hydrology / Biochemistry and Ecotoxicology; G. Reifferscheid, German Federal Institute of Hydrology, M. Wagner, Norwegian University of Science and Technology / Department of Biology. Microplastic (MP) toxicity has been considered in numerous taxa including bivalves, which are of special interest due to their high filtration activity and therefore MP particle uptake. Previous studies in marine bivalves reported stress and inflammation processes in response to high levels of MP exposure, while data on freshwater species is missing. Therefore, we aimed to assess the effects of irregular polystyrene MP (< 63 μm) on the lipid peroxidation and antioxidant capacity in the freshwater bivalve *Dreissena polymorpha* both in a single and multiple stressor exposure regime. We exposed *D. polymorpha* to polystyrene MP at concentrations between 6.4 and 100,000 µM for 6 weeks at 16 °C. After the exposure, the mussels were collected and analyzed for malondialdehyde concentrations as an indicator for lipid peroxidation (TBARS assay, thiobarbituric acid reactive substances) as well as for the remaining abundance of hydrophilic, non-enzymatic antioxidant substances (ORAC assay, oxygen radical absorbance capacity) – an estimate of the remaining antioxidant capacity. The analysis of lipid peroxidation

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and antioxidant capacity did not indicate any increased stress levels in response to chronic MP exposure in *D. polymorpha*. In addition, the same experiment performed in a sub-chronic exposure (1, 3 and 7 d) did not reveal stress-induced effects either. Therefore, this study indicates that polystyrene MP does not induce a stress response in *D. polymorpha* in the current exposure scenario. In a more environmentally realistic scenario, bivalves will experience other stressors (e.g. increased water temperature) besides particulate matter. Thus, we hypothesize that a different stress response can be modulated by MP exposure. To explore such a scenario further, we will present results from ongoing multiple-stressor experiments in which we expose *D. polymorpha* to MP at 16, 24 and 28 °C.

**TU183 Tissue Translocation of Polystyrene Micro- and Nanoparticles in Daphnia magna**

C. Schueg, Goethe University Frankfurt / Dpt. Aquatic Ecotoxicology; S. Rist, DTU, (Technical University of Denmark) / Department of Environmental Engineering; N.B. Hartmann, Technical University of Denmark (DTU) / DTU Environment; M. Wagenius, Norwegian University of Science and Technology / Department of Biology

The last decade has seen a surge in research investigating various aspects of micro- and nanoplastics originating from plastic pollution in aquatic ecosystems. Aspects include occurrence, uptake, and potential effects in biota. Working with particles in a laboratory setting bears its own kinds of challenges, some of which have already been faced by researchers in the realm of nanotechnology. Our elucidation of biota-particle-interactions is still limited and often based on early studies that – due to the infancy of the field – may have deficiencies in the experimental design and quality controls. One such example relates to the potential of plastic particles to cross the gut epithelium and translocate to other tissues. This phenomenon has been reported in the literature for the freshwater cladoceran *Daphnia magna* and – if true – may be a major route for toxic substances. To substantiate the limited available data, the aim of our study was to replicate these findings. We exposed neonate daphnids in a number of scenarios regarding particle concentration and exposure duration at two independent geographical locations using animals from two separate cultures. We expanded on the previous experiment by improving imaging through the addition of a fractoscope-based clearing followed by investigation through confocal laserscan microscopy. We additionally applied the lipophilic dye Nile red to localize lipid droplets. This step facilitated the identification of lipid droplets inside the tissue and could therefore associate fluorescence detected before staining to a respective tissue. Our findings potentially challenge previous publications that reported the translocation of both micro- and nanoparticles. This discrepancy may be based on false-negative results on our side or false-positive results in the earlier reports, both potentially caused by inadequate exposure settings during the investigative parts of the studies. We were unable to replicate these findings implying a tissue translocation of nano- and microplastics under conditions closely resembling those reported in the literature. This highlights that the replication of nano- and microparticles studies is important, especially if these have a high impact on the body as indicated in earlier macro-scale studies. In this study, we hypothesize that inhibition by a lack of transparency in reporting methodology and results. We were able to adapt a fractoscope-based clearing protocol to the use with high amounts of *Daphnia* samples.

**TU184 Do terrestrial organisms, isopods Porcellio scaber and earthworms Eisenia Andrei, avoid microplastic contaminated soil?**

A. Jemec Kokalj, University of Ljubljana, Biotechnical Fac. / Department of Biology; P. Židar, University of Ljubljana / Department of Biology Biotechnical Faculty; G. Kalčikova, University of Ljubljana / Faculty of Chemistry and Chemical Technology

Microplastics (MP) can potentially enter the terrestrial environment via sewage sludge deposition on agricultural land. In some countries plastic bags are used as soil cover in home gardens and agricultural land to act as mulch. Due to fragmentation of these plastic bags, secondary MP may enter the soil and can be further transported along the soil column by bioturbation. Despite the potential presence of MP in the terrestrial environment, data regarding the effects of MP on terrestrial organisms are very scarce. In this study, we investigated if terrestrial isopods *Porcellio scaber* and earthworms *Eisenia andreii* avoid soil contaminated with microplastic. We tested microplastic extracted from facial scrub and from sewage sludge with soil by flow cytometry and viSNE, which allows the 2D clustering of particles with different features according to the fluorescence measured. Exposure experiments were carried out for up to three weeks, using different types of microplastic particles and a wide concentration range. In the flow, when particles were mixed with fish tissue, flow cytometry/viSNE was able to differentiate particles natures, numbers and sizes. About 10% of added particles were internalized by the fish from all particles that floated or settled on the bottom. Particles ingestion resulted in a slight impact on behavior. Yet, floating particles were massively incorporated by the fish and significant numbers remained even after 24 h of depuration. Based on this, we are currently exploring if continuous...
feeding with microparticles contaminated food has consequences on juvenile fish growth. Taken together, our study demonstrates the power and limits of flow cytometry/viNSF for microplastics quantification in a complex biological matrix like fish. The setup could be extended to other types and forms of microplastics in different environmental matrices. Moreover, our study sheds light on ecological consequences that microplastics exposure might have on fish.

**TU187**

**Microplastic ingestion by fish: a comparison of Thames Estuary and Firth of Clyde populations**

A.R. McGorran, Royal Holloway; P.R. Cowie, Field Studies Council Scotland; P.F. Clark, The Natural History Museum; J.P. McEvoy, D. Morriss, Royal Holloway Thames Estuary and Firth of Clyde compare the ingestion of microplastic by pelagic and benthic fish populations from two major UK watersheds: the Thames Estuary and the Firth of Clyde. A total of 760 fish from 20 species and 116 brown shrimp, *Crangon crangon*, were sampled. Individuals were examined under a dissecting microscope and potential plastics were removed to be later identified by FTIR analysis. Out of 21 species, including both fish and shrimp, sixteen species from different trophic levels ingested plastics. Overall, between 33–47% of fish ingested plastics mostly fibres (85% of potential plastics; before FTIR analysis). In addition, microplastics were also found in the stomach of a common prey species, *C. crangon*, but had ingested far less plastic than predatory fish species, such as the European flounder, *Platichthys flesus*. In the Firth of Clyde, benthic, flatfish ingested significantly more plastic than pelagic fish and other benthic fish. This may indicate that, in estuarine systems, plastics accumulate in the sediment.

**TU188**

**Polystyrene microplastic uptake and effects on feeding behaviour and reproduction in the cladoceran Daphnia magna**

B. De Felice, Università degli Studi di Milano; R. Bacchetta, University of Milan; P. Trémolières, University of Milan / Department of Biomolecular Sciences and Biotecnology; M. Parolini, University of Milan / Department of Environmental Science and Policy

Plastic contamination is a well-known environmental problem as demonstrated by the huge presence of plastic debris ranging different sizes in diverse aquatic ecosystems worldwide. In recent years, the attention has been attracted to microplastics (MPs), small plastic particles (dimensional range Daphnia magna) affecting food intake, growth and reproduction. First, we performed a 24 hours uptake and 48 hours release test to assess the ingestion and elimination rate of MPs in daphnids. Already after 1 hour of exposure we found that MPs fill up the digestive tract of daphnids at all the tested concentrations. On the other hand, release test after 24 and 48 hours of exposure of MPs were still found massively in the digestive tract of treated individuals. The lack of a complete release of MPs can cause the blockage of the digestive tract and starvation, leading the crustacean to the death. Moreover, these effects can negatively affect body growth, swimming activity and, consequently, have strong consequences on reproduction, as suggest by a standard 21-day reproduction test.

**TU189**

**Uptake of differently sized microplastics in gut passage by different species of Daphnia**

S. SUPIAN, University of Birmingham; I. Lynch, University of Birmingham / Geography Earth Environmental Science; J. Sadler, The University of Birmingham / Earth Environment Science

Plastic-debris, for example from textile weathering and washing, are increasingly accumulating in aquatic environments, and while they are now recognized as environmental pollutants, their impact on aquatic ecosystems is not yet fully understood (Jemec, 2016). Microplastics, which are synthetic polymers with a diameter smaller than 5 mm and extending down to the nanoscale, have a widespread occurrence and negative effects on different trophic levels have been described (Hollman, 2013). The freshwater crustaceans Daphnia inhabit all kinds of aquatic systems and forms part of the plankton community acting as algae grazers (filtrators) and predators (raptors) making them an important indicator species in the foodchain. The Daphnia family includes species ranging in size from D. magna (2.5–3 mm) to D. galeata (1.3–2.0 mm) which spans a similar range of sizes as micro and nanoplastics, thus suggesting that different members of this family may be differentially sensitive to or affected by different sizes of micro or nano plastics. This work presents a first analysis of the effect of Daphnia body and gut size on uptake of microplastics of different sizes. We investigated the ingestion and effects of polybead carboxylate microparticles (0.1, 1.0 and 10 μm) on freshwater cladocera of different body sizes (D. magna, D. pulex and D. galeata) after 24, 48, and 72 hours exposure to a range of mass concentrations (also considered on the basis of particle number) (Nasser, 2016). The hypothesis tested was that the size of microplastic particles preferentially taken up by the organisms will scale with organism size, due to differences in their gut sizes and filter feeding capabilities. We assessed the uptake, accumulation, and depuration of the microplastics in Daphnia species using stereomicroscope (Nikon SMZ800) measurements. Image analysis approaches (ImageJ and Matlab) were utilised to calculate gut area, and pixel density within the gut, in an effort to quantify particle uptake. This was correlated with fluorescence measurements using fluorescently labelled microplastics, corrected for the average fluorescence per particle. Rapid accumulation in the gastrointestinal tract was observed after exposure to all particle sizes in D. magna, with the smaller particle sizes being detectable in the guts of neonates of all three species. The comparative uptake and effects of polybead microplastics on D. magna, D. pulex, and D. galeata are presented here for the first time.

**TU190**

**Determination of microplastics in mackerel stomachs by enzymatic digestion and μFTIR**

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Plastic is one of the most used materials in the world and is one of the most common and persistent pollutants in the oceans. In fact, plastics constitute 60-80% of marine litter. A particular fraction of plastic debris are the microplastics (0.5-5 mm). The presence and accumulation in the ocean is cause for concern for several reasons, one of the most important is that they can be ingested by marine biota [1]. Different studies have shown the effects on the biota, such as intestinal blockage, decreased mobility or death [2]. Microplastics can absorb persistent bioaccumulative and toxic compounds from seawater. Once ingested, the absorbed pollutants may be transferred to the respective organisms. A variety of methods has been developed to measure microplastics in biota. One important aspect of these analytical methods is the extraction of microplastics from interfering biomass. Many studies have employed one or more chemicals (KOH, H2O2) to dissolve the biomass, which can be destructive to the plastic particles and their surfaces and create interferences that were problematic for μ-spectroscopy-based analyses. Enzymatic digestion methods have been used to minimize damage to plastics [3]. An enzymatic digestion has been developed and optimized for digesting biological material without destroying microplastics. Different times and enzymes were tested to optimize the enzymatic protocol. In addition, the enzymatic protocol was compared with chemical digestion (KOH) for the treatment of mackerel stomachs. The optimized enzymatic protocol has been applied to quantify the microplastics debris present in the mackerel stomach. Identification and characterization of microplastics was done by μFTIR. **Acknowledgement**: Financial support is acknowledged to the Program of Consolidation and Structuring of Units of Competitive Investigation of the University System of Galicia (Xunta de Galicia) potentially co-financed by ERDF (ED443IC170278) and by the Ministry of Economy and Competitiveness (subproject CTS-2015-170-C01-01-Fundos BASPEN (JPI Oceans) and, project CTM2016-77945-C3-3-R (ARPA-AUCA). **References**: [1] V. Hidalgo-Ruz, L. Gutov, R.C. Thompson and M. Thiel, Environmental Science & Technology 46, 3060 (2012); [2] M. Cole, H. Webb, P. K. Lindeque, E.S. Fleming, C. Halsband and T. S. Galloway, Scientific Reports 4, 4528 (2014) [3] J. Wagner, Z-M. Wang, S. Ghoshal, C. Rochman, M. Gassel and S. Wall, Anal. Methods, 9, 1479 (2017)

**TU191**

**Microplastic contamination of the model system Weser-National Park Wadden Sea: an across-ecosystem approach**

S. Moses, University of Bayreuth / Animal Ecology I; L. Schrank, C. Lorforsch, University of Bayreuth / Environmental Science

For the first time worldwide, in the joint project PLAWES the pollution with microplastics of a large European river basin will be investigated on the example of the model system Weser-National Park Wadden Sea. PLAWES, as a pioneer study, is going to conduct an interdisciplinary and ecosystem overarching analysis concerning the contamination with microplastics from the headstreams to the North Sea, thereby considering exemplarily major point (e.g. wastewater treatment plants, combined sewer systems) and diffuse (drainage, atmosphere) sources and entry routes. The new insights are going to be included in a new modeling concept for the identification of primary transport mechanisms and accumulation zones of microplastics. Effects of microplastics on ecosystems of the Weser-Wadden Sea system will be investigated on both, aquatic invertebrates and the interaction of pathogens with microplastics in biofilms. The insights on ecologically relevant aspects are going to be used to assess the environmental effects of microplastics on the model system Weser-National Park Wadden Sea and to transfer these to other systems. Furthermore, the results will be used to develop novel teaching materials to provide an education platform for teachers, pupils and parents across Europe. Hence PLAWES will generate scientific data on the impacts of microplastics on a large European river basin and on environmental health. This will not only be instrumental for decision makers and stakeholders but also serve as focal point to develop science-based solutions.

**TU192**

**Photochemical fragmentation of freshwater (micro)plastics under UV irradiations**

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We begin to understand and describe more and more the fate of a plastic waste arriving (and remaining) in the aquatic environment. Nevertheless, we still do not know many things, for example, the time scaling of the process from the abandonment of a waste, its arrival, and its persistence in the aquatic environment. During this period, the material will be exposed to various environmental aggressions that will initiate and spread the photoaging of the material. This scenario is accompanied by a physical fragmentation into microplastics of increasingly smaller sizes, and by a chemical functionalization due to the photo-oxygenation of the macromolecular chains. Finally, the increase in both the specific surface area and the chemical functionality may influence strongly the interaction parameters with persistent organic pollutants. We have studied, in simulated laboratory conditions, the fate of various plastic fragments (Polystyrene, Polypropylene and Polyactic Acid) immersed in fresh water and UV irradiated. We worked either with real wastes (from post-consumer sector) or with model polymers totally free of additives. The polymers were chosen for their different physical properties. Polystyrene behaves like a glass ($T_g = 104^\circ C$) at the temperatures of use, which is not the case of the PP ($T_g = 0^\circ C$). Finally, PLA can start hydrolysis reactions. During the exposure time, solids and liquids (a small volume of water) are taken for analysis (membrane filtration, ion and liquid chromatography). The main result is that in all cases there is a formation and leaching of short chain (1C, 2C, 3C) carboxylic acids (acetic and formic acids, lactic and glycolic in the case of PLA) all known to be markers of polymer degradation. In the absence of light no transfer of any acid in the liquid is recorded.

TU193
Characterization and Environmental Risk Assessment of Polymeric Cosmetic and Personal Care Ingredients
J. Davies, Personal Care Products Council / Science

Polymers have been developed to perform a variety of functions that are central to modern living. Cosmetic and personal care products (CPCPs) contain a wide array of polymeric ingredients which are identified by the International Nomenclature of Cosmetic Ingredients (INCI). An INCI name often represents several polymers with different physical and chemical properties. This often leads to one INCI named polymer existing as several physical forms. For example, polyethylene can exist as a solid plastic microbead or a non-plastic wax thickening agent. The presence of polymers in the environment, particularly plastics, is of growing concern, yet relatively little is known about the environmental risk these materials may pose or how this can be assessed. The CPCP industry therefore developed a risk-based prioritization framework for polymeric ingredients. Polymers are characterized by their physchem properties. Solid polymers, such as plastics, are prioritized for assessment since they are routinely detected in the aquatic and marine environment and thus have the potential to contribute to the litter load. Toxicity is that in exposure and hazard of priority polymers is then assessed. When a polymer poses an unacceptable environmental risk, risk mitigation options are considered. A polymer’s physchem properties provide insights into how environmental exposure and hazard can be assessed. A decision tree was developed linking physchem properties to methods for assessing polymers. Depending on a polymer’s properties and how these may change in the environment, polymers are either assessed following existing polymer assessment guidelines (such as those prescribed by USEPAl) or use of novel methods. The work presented provides a scientifically robust approach for accurately assessing the impact of polymers in the environment.

TU194
Toxicological effects of irregularly-shaped and spherical microplastics in a marine teleost, the sheepshead minnow (Cyprinodon variegatus)
J. Park, Korea Institute of Toxicology; J. Choi, Korea Institute of Korean KIT; Y. Jung, Korea Institute of Toxicology; S. Hong, Korea Institute of Ocean Science and Technology / Oil and POPs research group

Increasing worldwide contamination of the marine environment with plastics is raising public concern of potential hazards of microplastics to environmental and marine organisms and particularly in marine ecosystems. Sediments are known as the most important contaminated environmental compartment. Thus, the aim of this study was to assess the MP contamination in sediment from the French Atlantic coast. Sediments were sampled at three locations (Payes de la Loire region, France) and in two seasons: October 2015 (beginning of autumn) and March 2016 (beginning of spring). Firstly, different protocols (with or without digestion step) were tested according to the literature and the most appropriate was validated spiking 23 g of sediment samples in triplicates with 4 different MP types: polyethylene (PE), polypropylene (PP), polyvinyl chloride (PVC) and polyethylene terephthalate (PET). MP extraction from sediments was performed using milliQ-water combined with centrifugation technic. After a filtration step, MPs were detected and identified directly on the membrane filters using µFTIR spectroscopy in reflection mode. Then, a test was performed in order to determine the replicate number required to obtain a celluly representative of the whole sampled sediment. For the sediment collected in the field, MPs were found in each location and for each season. Average levels ranged from 38 (± 46.72) to 102 (± 105.37) MP per kg of dry sediment (N = 10; 250 g). Ten different compositions of MPs were defined by µFT-IR with a high proportion of PE and PP, 38 and 23% respectively. Five MP types (PE, PP, PVC, polystyrene and polyester) represented more than 90% of MPs. Interesting information of MP characteristics supported the explanation of the source and also the long-time passed in marine environment. None of significant differences were found among six sample groups. This work provides the first dataset on the level of contamination in sediments from the French Atlantic Coast.

Derivation, Validation and Implementation of Environmental Quality Benchmarks (P)

TU196
Challenges in implementing legal frameworks for assessing water quality: the cases of the EU and Swiss approaches
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Human activities have a great impact on river quality. Monitoring programs show that multiple chemicals are present in water and that physico-chemical properties and runoff/dissolution capacity of rivers evolve due to climatic changes. These changes can affect the aquatic ecosystems as well as the amount of useable water. It is therefore crucial to evaluate the state of river systems using a holistic approach. The European Commission established a framework to highlight rivers’ ecological deficits and to enhance regional or local water management plans. In Switzerland, such a framework is currently under development. In this study, we compared both procedures and implemented them in a Swiss catchment dominated by agricultural activities. The goal was to identify challenges linked to the application of these approaches. Both frameworks consider the idea that no single assessment alone meets a good environmental state and that the latter deteriorates as tributaries and wastewater discharge flow into the main riverbed. Chemical issues and water quality changes due to hydro-climatic variations and management strategies were also pinpointed. Both frameworks are thus able to highlight the main problems of the river and are consistent with each other. They are thus useful tools to survey the spatial and temporal evolution of rivers quality. However, several challenges remain, especially regarding the strategies to monitor and analyze chemicals, the definition of target values and conditions, the evaluation and integration of human-induced-pressures, and the overall evaluation of the rivers state. Development of integrated indicators is seen as a potential solution to explore river health and to provide efficient restoration measures by water managers.

TU197
Updating the Environmental Quality Standards for the EU priority substance chlorpyrifos
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Chlorpyrifos (CPF) is widely used as an active ingredient in insecticides. Since 2005 CPV is a priority substance under the EU Water Framework Directive (WFD) with an AA-EQs of 0.033 mg/L and a MAC-EQs of 0.1 mg/L. The aim of this study is to update the Environmental Quality Standards (EQS) for chlorpyrifos in the current data and the WFD method for EQS derivation published in 2011. Both AA-EQs and MAC-EQs decreased by more than one order of magnitude. The original AA-EQs was not derived based on available chronic ecotoxicity data but was set as MAC-EQs divided by a factor of 3, while the revised value of 0.00046 mg/L is based on a NOEC for A. bahia taken from the EESA authorisation dossier and was an assessment factor (AF) of 10. The original MAC-EQS was derived from mesocosm NOECs using an AF of 1. The revised MAC-EQS of 0.0044 mg/L is based on an HC, from a species sensitivity distribution (SSD) for crustaceans and insects using
Lead exposure in European Freshwaters: are they a risk? A regulatory assessment accounting for bioavailability

I. Wilson, A. Peters, G. Merrington, wca; J. Chowdhury, International Lead Association / Senior Scientist -Environment

Lead (Pb) is a chemical for which one EQS has been set and is applied across all continental water bodies in the European Union. The EQS (Environmental Quality Standard) is used in Europe is through a compliance assessment, effectively a comparison of the measured concentration of the chemical of interest in a water sample and the EQS. An indicative tiered compliance assessment of the European-wide bioavailable lead EQS of 1.2 µg L−1 (EQSbioavailable) was undertaken against regulatory freshwater monitoring data from six European member states and FOREGS database. A tiered approach comparing the measured metal concentrations to be compared for, by correcting the measured dissolved metal concentrations in the water sample to a bioavailability-based concentration to be compared to an EQSbioavailable. In Tier 1 measured concentrations were compared against the EQSbioavailable. In Tier 2, Bio-net, a user-friendly tool based upon Biotic Ligand Models (BLMs) was used to account for bioavailability, along with the current European Water Framework Directive lead dissolved organic carbon correction (DOC-WFD) approach. The outputs from both approaches were compared to the Biotic Ligand Model (BLM), the final tier in the tiered approach. Lead exposures are relatively low across all regulatory datasets and FOREGS. At Tier 1, only 3.9% of sites and samples assessed have a dissolved Pb concentration of greater than or equal to the EQSbioavailable. The waters showing the greatest sensitivity to potential lead exposures are characterized by relatively low DOC (< 0.5 mg L−1), regardless of the pH and calcium concentrations. Whilst some risks due to lead are possible this is due to the combination of sensitive waters with elevated lead concentrations, as there is very little overlap of the distributions of site specific PNEC values and dissolved exposure concentrations.

Assessing compliance of European Freshwaters for copper: accounting for bioavailability

A. Peters, I. Wilson, G. Merrington, wca; D. Heijenrickx, ARCHE; S. Baken, European Copper Institute

The importance of accounting for bioavailability in understanding the effects of metals has long been recognised in terms of setting environmentally relevant regulatory limit values. Attempts to deliver practical, routine methods to do so have been limited. For regulatory purposes in Europe an agreed Environmental Quality Standard (EQS) for a substance must be a fixed value: the same EQS is applied for all jurisdictions and geographic areas, irrespective of the concentrations of metals, the EQS must be set for the bioavailable forms, and it is termed EQSbioavailable. This study determines the levels of compliance of European freshwater with a copper EQS, and evaluates the usefulness of a tiered approach to compliance assessment for copper. The first tier compares the dissolved metal concentration to a threshold, estimated using either regional or continental water chemistry data. At Tier 2, the bioavailable metal concentration is calculated using the physico-chemistry of the water body, and compared to the EQSbioavailable. It follows that the thresholds at Tier 1 must be set at a level which will ensure protection of sensitive environments. The value of the threshold has important implications in terms of the financial costs of the compliance assessment. For copper, setting the thresholds at the same level for the whole of Europe (i.e. continental) would leave some countries with costly and unnecessary monitoring requirements. Deriving the threshold on a continental basis is also country specific basis enables effective use of resources without compromising on the level of protection. A very high level of compliance for copper is observed where bioavailability based thresholds are used for the implementation derived from regionally relevant water chemistry data (99.3%). Sites where elevated ambient background levels of copper are combined with very high bioavailability, principally when the waters have low DOC concentrations, are those most likely to be at risk due to copper exposures.

Are lead exposures a risk in European Freshwaters? A map of EQS compliance assessment accounting for bioavailability

J. Chowdhury, International Lead Association / Senior Scientist -Environment; A. Peters, I. Wilson, G. Merrington, wca

Lead (Pb) is a priority substance for which the bioavailable Environmental Quality Standard (EQSbioavailable) of 1.2 µg L−1 has been set under the European Commission directive 2013/50/EU for application across all countries in Europe. In the present study, a Tiered approach was applied to undertake a compliance assessment of the EQSbioavailable using the FOREGS database that includes paired data for water quality parameters and measured Pb concentrations from freshwater streams and rivers across Europe. In Tier 1 measured dissolved Pb concentrations were directly compared against the EQSbioavailable. In Tier 2, Bio-net, a user-friendly tool based upon Biotic Ligand Models (BLMs) was used to account for bioavailability, along with the current European Water Framework Directive lead dissolved organic carbon correction (DOC-WFD) approach. The outputs from both approaches were compared to the chronic Pb Biotic Ligand Model (Pb BLM), the final tier in the tiered approach. The maximum Pb concentration in FOREGS is approximately 11 µg L−1. At Tier 1 screening, only 16 (2.0%) water samples of the whole dataset (n=797) have Pb concentrations that are greater than the EQS of 1.2 µg L−1. The exceedances further decreased to 3 (0.4%) and 1 (0.1%) upon accounting for bioavailability at Tier 2 and 3 respectively. The map of site-specific PNECs (predicted no effect concentrations) as calculated by the Pb BLM identifies that the most sensitive waters in the database are all those with extremely low concentrations of DOC (< 0.5 mg L−1). The greatest frequencies of such sites are found in the Alps and Norway. The lowest PNEC values for Pb are around 0.5 µg L−1, and the WFD EQS value of 1.2 µg L−1 is equivalent to approximately the 4th percentile of the dataset. The results indicate that the European freshwater bodies with low anthropogenic pressure are unlikely to fail the compliance with the EQS, with the exception of very local situations such as historic mining sites.

Integrated approaches in ecotoxicology: bridging the gap between experimental toxicology and mechanistic modelling (P)

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Testing, analysing and predicting the lethal action of chemicals on organisms plays a central role in the fields of ecotoxicology and toxicology, both for scientific and regulatory purposes. The dominant approaches to deal with survival data are descriptive, focussing on standardised tests and simple summary statistics (such as the LC50). Such descriptive methods ignore the fact that lethal effects develop over time, thereby leading to biased assessments and precluding useful predictions to untested exposure scenarios. Making sense of toxic effects over time requires mechanistic models, and, more specifically, the explicit consideration of toxicokinetics and toxicodynamics (TKTD). For the endpoint survival, almost all existing TKTD models can now be viewed as special cases of a mechanistic modelling framework: GUTS, the General Unified Threshold model for Survival. GUTS was conceived in 2010, and has since gradually been a large user community. Furthermore, the model is receiving increasing interest from the regulatory field as it is explicitly suited for the analysis of survival data, and for extrapolation across different exposure scenarios. With the increasing interest in GUTS, and the increasing interest in good-modelling practice, it is time for a more detailed treatise on this model framework. In a CEFIC-LRI funded project, we have prepared an extensive e-book on GUTS (which will be available for download, free of charge, January 2018). The book contains a detailed description of the model framework (concepts, underlying assumptions and mathematics) and its historical roots, as well as worked-out case studies, guidance for users of the model (or its results), and the results of a ring test for a range of software implementations. This book is the standard work on GUTS modelling, and the e-book format allows the contents to be kept up to date with the major novel developments in this area.

Ecosystem response modelling in aquatic and terrestrial effect models

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In recent years mechanistic effect models including GUTS and DEBTox have been successfully used in the aquatic and terrestrial risk assessment of pesticides. These models offer the advantage that results from laboratory studies, usually conducted with constant exposure, can be extended to time variable exposure, which is more typical under field conditions. At present these models consider a threshold beyond
which effects start to appear. Once this threshold is surpassed the amount of effect is calculated using a linear regression, i.e. effects increase linearly with increasing concentration. In other areas of the risk assessment dose-response modelling and the derivation of reliable dose-response curves has received much focus (see e.g. the new EFSA guidance on benchmark dose modelling). Therefore, it is investigated in the present study if these models can be improved by considering more typical dose-response curves, which often are sigmoidal shaped. It is investigated when the specific shape of a dose-response curve affects the outcome of an assessment and how the magnitude of predicted effects is affected.

**TU 203 Investigating toxicokinetics of emergent pollutants (PFASs) in the common sole (Solea solea) from in situ measurements and experimental data on PCBs within a DEB-based modelling approach.**

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In the context of global change, developing mechanistic tools integrating the influence of environmental factors on toxicants bioaccumulation dynamics is required, as organisms will face unprecedented conditions. Based on the Dynamic Energy Budget (DEB) theory, it is possible to predict individual and population level consequences. However, for emerging chemicals like Perfluoralkyl Substances (PFASs), toxicokinetics calibration is a challenge as their properties are not fully characterized yet and experiments are scarce. Here we present the results of a research project aimed at investigating toxicokinetic models (TKM) of PFASs in juvenile common sole from a contaminated nursery ground, the Gironde Estuary (France). In a highly fluctuating environment, our goal was to extract information from the large variability observed in the measurements for several polychlorinated biphenyls (PCBs) and PFASs. First, we designed environmental scenarios and prioritized environmental sources of inter-individual variability using a mechanistic model calibrated for CB153 thanks to experimental data. As CB153 is poorly biotransformable in fish, its bioaccumulation in juveniles mainly relies on the ingestion of contaminated food. Further, we considered this congener as an additional tracer of potential preys in the wild. Indeed, diet reconstruction from stomach contents and isotope data provided us with contrasted pictures. We proceeded jointly to: a soil food chain investigation for a range of environmental diets, food contamination and temperature scenarios. Comparing these predictions with in situ measurements, we were able to highlight the major influence of diet composition. The next step was to consider other PCBs and PFASs with previously selected environmental scenarios. Discrepancies between model predictions and observations allowed us to formulate new modelling hypotheses taking into account the possibility of soil contamination and various halflives for different compounds and food density depending on their properties (e.g. hydrophobicity, spatial conformation, functional groups). This mechanistic approach prioritizing sources of variability provided new insights on the differential bioaccumulation between toxicant families in a key flatfish resource.

**TU 204 Investigating metabolic acceleration in dynamic energy budget models of copepods using the ecotoxicological model organism Nitocra spinipes**

J. Koch, GhEnToxLab (Ghent University) / Applied Ecology and Environmental Biology; K. De Schampaere, Ghent University (UGent) / Applied Ecology and Environmental Biology

Copepods form an essential part of marine ecosystems and constitute a large portion of animal biomass on earth. Moreover, their small body size and short life cycle make them convenient test organisms in ecotoxicity studies. Beside acute toxicity, multiple works in the past focused on chronic life history effects of chemicals in copepods. Unfortunately, we usually lack a mechanistic explanation of observed effects, and models for realistic lab and field studies are often rooted in Dynamic Energy Budget (DEB) theory to help evaluate sublethal toxicity data in terms of effects on the energy household of an animal. Although DEB models for new species are usually easily parameterised, the copepod life history shows distinct deviations from the ‘standard DEB model’ requiring further investigation. While some authors presume metabolic acceleration from birth until puberty, others suggest a von Bertalanffy growth curve which is truncated at the final molt. In this study we parameterised the two tyipified DEB models ‘abp’ (metabolic acceleration from birth to puberty) and ‘sbp’ (standard von Bertalanffy growth from birth to puberty) for the harpacticoid copepod Nitocra spinipes to investigate metabolic acceleration in copepods. As no high-quality data on length over time were available for N. spinipes, we performed a growth experiment over 28 days. Additional data from literature were used to aid the parameter estimation. Submodels for food (Holling’s type II functional response) and temperature dependency (Arrhenius temperature correction) were calibrated on development time and reproduction data. While isomorphic growth is commonly assumed in DEB studies, it does not hold true for N. spinipes which grows more slender in the course of its development. Hence, we used the square root of the top view area as a length measure to scale with the cubic root of structural volume in length-to-volume conversions. Both models abp and sbp showed good fits to the given data. Overall, abp predicted the data slightly better compared to sbp with a mean relative error of 0.063 vs. 0.076 in sbp. However, we do not regard this difference clear enough to unequivocally confirm or reject metabolic acceleration in copepods. More detailed data on N. spinipes and other copepods are needed to reveal the most accurate model for the copepod life history. That said, both models are promising tools for the evaluation and extrapolation of toxicity data in N. spinipes.

**TU 205 Grey seal physiology and environmental change**

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Main marine mammals are considered as sentinel species for marine ecosystem health. In the Baltic, grey seals (Halichoerus grypus) can serve this purpose as they are top predators and have shown to respond to anthropogenic and environmental stressors over the past decades. These stressors can influence the physiology and health of grey seals, ultimately leading to individual and population level consequences. Acknowledging the need for mechanistic understandings of stressor effects, we have developed a full lifecycle bioenergetics model for Baltic grey seals using Dynamic Energy Budget (DEB) theory. We use the comprehensive information available in the literature on grey seal physiology and metabolism and reproduction to parameterize and validate our model. Our model accurately predicted grey seal energy ontogeny and lifehistory traits, providing one of the first full descriptions of mammalian development in DEB. Recent reports have indicated that climate change effects on sea ice and food web dynamics have impacted grey seal condition (i.e. blubber thickness). We use our model to explore these relations and confirm the hypothesis that grey seal condition in the Baltic is susceptible to change in food quality/quantity and can lead to down-stream consequences on reproductive success. The results offer new insights into physiology and ecology of Baltic grey seals with the potential to lead to novel approaches for the study of stress ecology and conservation of this species.

**TU 206 Evaluation of thermal stress on Daphnia magna using oxidative stress and life-history trait parameters**

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Present study evaluated the effects of temperature (20 °C and 25 °C) on oxidative stress and life-history trait responses of Daphnia magna in short-term (5 days) and long-term (21 days) exposures. D. magna exposed to 25 °C exhibited continuous higher production of reactive oxygen species (ROS). In short term exposure, glutathione peroxidase (GPx) activity was significantly suppressed in elevated temperature. In contrast, daphnids showed significantly enhanced catalase (CAT) activity in long-term exposure. Glucose and lipid metabolism in the Baltic is considerably different as compared to that of temperate areas. Lower lipid peroxidation (LPO) level at elevated temperature under prolonged exposure suggests that antioxidant enzymes successfully prevented ROS-mediated damage. In addition, exposing D. magna to elevated temperature significantly shortened time to first brood, brood size, and body length, but induced significantly higher male production (p = 0.05). Reduced body length at elevated temperature indicates that D. magna may not be able to cope with temperature stress and reproduction to cope with the thermal stress. Moreover, a multi-generational study was performed to evaluate multigenerational effect on D. magna.

**TU 207 Transport-protein metal binding links uptake biodynamics for predicting copper in tilapia**

Y. Chen, Kaohsiung Medical University / Biomedical Science and Environmental Biology; W. Chen, Kaohsiung Medical University / Department of Biomedical Science and Environmental Biology; C. Liao, National Taiwan University / Department of Biomedical Science and Environmental Biology

Metal could bind to transport protein, then accumulate in the cellular and tissue. It points out that the metal ion accumulating in target subcellular compartment could reflect the metal toxicity. Copper (Cu) plays an essential role in cellular metabolism of aquatic organisms, but it would cause toxicity with excessive accumulation. The purpose of this study was to conduct the short-term exposure experiment to examine the Cu accumulation in tilapia, then combined with bioavailability and subcellular partitioning to estimate the Cu binding situation and mechanism of toxicity on gill. We developed a mathematical framework that quantified the Cu affinity and the amount of transport protein in different subcellular compartment. Results indicated that Cu accumulation in metabolically active pool (MAP) was preferred to organelles than heat denatured protein, and Cu accumulation in metabolically detoxified pool (MDP) was metal rich granule. The estimated parameters of maximum Cu influx rate, total number of transport protein and affinity constant didn’t have significant differences between MAP and MDP. However, the conditional stability constant of MDP 0.45 ± 0.005 μl g⁻¹ was

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significant higher than that of MAP 0.269±0.018 mg μl⁻¹ (p < 0.001), and the uptake rate constant of MDP 0.128±0.001 ml g⁻¹ h⁻¹ also was significantly greater than MAP 0.086±0.001 ml g⁻¹ h⁻¹ (p < 0.001), that revealed that Cu was likely to bind on MDP in the low exposure concentration than that of MAP. This study concluded that Cu tend to accumulate in MDP, then may cause less toxicity to tilapia. Keywords: Copper; bioavailability; transport protein; subcellular partitioning

TU208
Relationships between subcellular metal partitioning and biomarkers of effects in white suckers (Catostomus commersonii) exposed to an environmental metal gradient
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Discharges from metal mining operations may lead to metal accumulation and toxicity in aquatic species. Once metals enter cells, they can bind to sensitive components and cause deleterious effects. Nevertheless, metals can also be detoxified by binding to nucleotides designed to sequester them, limiting their toxicity. The objectives of this study were (i) to assess the subcellular distribution of metals and metalloids (Cd, Cu, Se and Zn) in layers of white suckers exposed to metal-mining effluents, and (ii) to investigate the links between the binding of specific metals to particular subcellular fractions and physiological effects. To this end, mature male and female fish were collected in three lakes downstream from a metal-mining effluent and one lake in a reference area. Subcellular partitioning among putative metal-sensitive fractions (MSF) and biochemically detoxified fractions (BDM) in layers was determined after differential centrifugation and heat-denaturation steps. In parallel, a suite of biomarkers was investigated ranging from general indicators of energy accumulation to specific indicators of oxidative stress and metabolic or biosynthetic capacities. Total hepatic metal concentrations were significantly higher in exposed fish than in reference fish, with Cd in the x10 and Se (x10) being accumulated the most. No differences between sexes were observed. Subcellular partitioning of metals was similar among areas but specific to metals; over 70% of the Cd burden was found in the heat-stable cytosolic proteins fraction, which includes metallothionenins. In contrast, the largest contributors to the total Se liver burden were the potentially metal-sensitive heat-denaturable proteins fraction (=35%), and the organelles fraction (=30%). These results suggest that Cd was well detoxified and regulated by white suckers, whereas the presence of relatively high Se concentrations in the MSF suggests that exposed fish were likely subject to stress. Principal component analysis showed that increasing [Se] in all of the fractions was strongly correlated with lower fish condition and associated with higher concentrations of a trend in Se concentration in the fish body, and Se in fish muscle activation. Acetylcholinesterase (AChE) is an enzyme that hydrolyzes acetylcholine in order to eliminate it from the body, and when AChE is inhibited acetylcholine levels increase. Excess acetylcholine at cholinergic synapses overstimulates muscarinic- and nicotinic-type receptors. These receptors are found in most organs of the body, thus multiple adverse outcomes may result. Moreover, a wide variety of chemicals including organophosphates, carbamates and some high nitrogen compounds, can inhibit AChE. Thus, the impact of AChE inhibition is large, yet relatively little research has been focused on developing related adverse outcome pathways (AOPs) or a network for this molecular initiating event. This presentation focuses upon the construction of adverse outcome pathways that result from AChE inhibition in zebrafish (Danio rerio). We performed a comprehensive literature review of the literature to identify studies and datasets that could be used to construct an AChE AOP. Adverse outcomes include seizures, impairment of the retina architecture and behavioral changes. Preliminary AOPs for these outcomes will be presented with references to the studies that support the AOP, and identification of data needed for quantitative AOP development.

TU209
Development of an adverse outcome pathway for acetylcholinesterase inhibition in zebrafish (Danio rerio)
K.H. Watanabe, A. Mikhail, Arizona State University / School of Mathematical and Natural Sciences; K. Conrow, Arizona State University; N. Vinas, Mississippi State University / Engineer Research and Development Center

Acetylcholine is a neurotransmitter that is important for a broad range of processes in mammalian body such as muscle activation. Acetylcholinesterase (AChE) is an enzyme that hydrolyzes acetylcholine in order to eliminate it from the body, and when AChE is inhibited acetylcholine levels increase. Excess acetylcholine at cholinergic synapses overstimulates muscarinic- and nicotinic-type receptors. These receptors are found in most organs of the body, thus multiple adverse outcomes may result. Moreover, a wide variety of chemicals including organophosphates, carbamates and some high nitrogen compounds, can inhibit AChE. Thus, the impact of AChE inhibition is large, yet relatively little research has been focused on developing related adverse outcome pathways (AOPs) or a network for this molecular initiating event. This presentation focuses upon the construction of adverse outcome pathways that result from AChE inhibition in zebrafish (Danio rerio). We performed a comprehensive literature review of the literature to identify studies and datasets that could be used to construct an AChE AOP. Adverse outcomes include seizures, impairment of the retina architecture and behavioral changes. Preliminary AOPs for these outcomes will be presented with references to the studies that support the AOP, and identification of data needed for quantitative AOP development.

TU210
Development of a Novel Quantitative Adverse Outcome Pathway Predictive Model for Lung Cancer
T. Hill, US EPA NHEERL/ISTD/CB / ORD NHEERL Integrated System Toxicology Division; R. Conolly, US EPA RTP

Traditional methods for carcinogenicity testing are resource-intensive, retrospective, and time consuming. An increasing testing burden has generated interest in the adverse outcome pathway (AOP) concept as a tool to evaluate chemical safety in a more efficient, rapid and effective manner that better directs resource utilization. A central premise of the AOP concept is that pathway progression from the molecular initiating event (MIE) implies a definable "response-response" (R-R) relationship exists between each key event (KE) that drives the pathway towards the adverse outcome. Computational description of these R-R relationships in a quantitative AOP (qAOP) enables dose-response consideration of probabilities and uncertainty, as well as flagging of special at-risk populations or sentinel species. The qAOP also provides a platform to utilize early genomic and in vitro data streams for rapid, less resource-intensive predictive models as well as the development of a computational approach that informs the level of risk. This poster describes a novel AOP/qAOP for lung cancer in the mouse from the MIE of CYP2F2-specific formation of reactive metabolites, advancing through KE for protein/nuclear acid adducts, diminished CC10 capacity and hyperplasia of CC10 deficient Clb cells, and culminating in the adverse outcome of mixed-cell type lung tumors in the mice. The qAOP is independent of route of exposure and grounded in overlapping mechanistic events for naphthalene, styrene, ethyl benzene, isoniazid and fluensulfone in the mouse. The qAOP modeling is supported by defined mechanistic relationships and quantitative data (PB-PK, dose-response and time-course) from archival data in peer-reviewed literature. Findings will include evaluation of data supporting the cancer qAOP, suitability for characterization of R-R relationships, and identification of data gaps or additional research as required. This approach supports international efforts on use of quantitative effect thresholds for adversity predictions and incorporation of novel data streams into the cancer risk assessment process. This abstract does not necessarily represent the views or policies of the U.S. EPA.

TU211
A combined PBTK and qAOP/modeling approach to assess the impact of dioxin-like compound (DLC)-induced embryo toxicity on recruitment failure in European eels
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The panmictic stock of the European eel (Anguilla anguilla) has seen a dramatic decline over the past several decades, and declines in recruitment as a result of maternally transferred contaminants has been proposed as one of several potential causes. In particular, dioxin-like chemicals (DLCs) have been identified as a class of chemicals of great concern for both European and American eels (Anguilla rostrata). DLCs bioaccumulate, are highly embryotoxic in many species of fish, and maternally transferred in artificially matured eels. However, to date researchers were unable to locate correlations with the birth eels in their natural spawning grounds in the Sargasso Sea. As a result, accurate embryotoxicity data to identify the potential causative chemicals are unavailable. Therefore, this study aimed to (a) parameterize a physiologically-based toxicokinetic (PBTK) model for European eels to account for the impact of changes in physiology that result from sexual maturation and migration on toxicokinetics, and (b) to couple this model with a quantitative adverse outcome pathway (qAOP) for activation of the aryl hydrocarbon receptor 2 (AHR2) of fishes to predict early life stage mortality of eels as a result of exposure to maternally transferred DLCs. The PBTK model was used to kinetically predict the redistribution of DLCs within the body of female eels during migration, and ultimately the concentration in gonads and eggs. A simple qAOP was described previously linking activation of species specific Ah receptor (AHR2) gene expression in vitro in eel hepatocytes to embryonic mortality in vitro. This qAOP was based on cell death (Annexin V positive) and embryo lethality across nine species of fishes exposed to DLCs. To this end, AHR2 was cloned from European eel and used to predict eel-specific relative potencies of five DLCs representing congeners measured at among the greatest concentrations in gonads of eels. Using this data, mortality of early life stages of eels was estimated based on the internal concentrations predicted by the PBTK model. Our integrated PBTK model and qAOP approach will ultimately shed light on the question whether early life stage mortality induced by exposure to DLCs has the potential to significantly contribute to the observed decline in recruitment of eels.

TU212
Salmonid pituitary cells as a test system for identifying endocrine disrupting compounds
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The pituitary gland is a glandular endocrine system, producing two gonadotropins, follicle-stimulating hormone (FSH) and luteinizing hormone (LH), which regulate gonadal development, sex steroid synthesis and gamete maturation. Despite its central role in regulating reproduction, there are limited data on impacts of endocrine disrupting chemicals (EDCs) on the pituitary gland. We have previously observed that waterborne exposure of previtellogenic coho salmon to 17β-ethynylestradiol (EE2) causes widespread effects on the pituitary transcriptome. Other in vivo studies with the selective serotonin reuptake inhibitor (SSRI) fluoxetine caused a decrease in FSH beta subunit (fsb) mRNA levels. These results motivated us to expand our studies by developing an in vitro test
system using pituitary cells isolated from coho salmon and previtellogenic female rainbow trout. Preliminary studies were performed to optimize culture conditions and to establish the time course of fshb and LH [lhb] subunit gene expression with and without addition of endogenous sex steroids (estrogen [E2] or 11-ketotestosterone). These initial studies suggested culturing with and without E2 was valuable as it could mimic the (+) feedback effects on LH that is observed from in vivo studies. After optimizing assay conditions, a suite of 12 contaminants and other hormones was evaluated for their effects on fshb and lhb gene expression. Each chemical was tested at 4-5 different concentrations up to solubility limitations in cell culture media. Results indicated more chemicals altered LH synthesis than FSH. The more potent chemicals were estrogens (E2E2) and aromatizable androgens (testosterone), which induced lhb. The estrogen antagonist 4-OM-tamoxifen decreased the E2-stimulated expression of lhb. Among several SSRI-tested, the sertraline metabolite norsertraline was notable for both increasing fshb synthesis and decreasing the E2 stimulation of lhb. These results indicate that diverse types of chemicals can alter gonadotropin production in fish. Further, we have shown that pituitary cell culture is useful for screening chemicals with potential endocrine disrupting activity and can support quantitative adverse outcome pathway testing. Supported by EPA-STAR grant R835167.

TU213 SETAC Mechanistic Effect Models for Ecological Risk Assessment of Chemicals Interest Group
E. Zimmer, IBACON GmbH

Integrating life cycle approaches towards a sustainable circular economy (P)

TU214 Metal and mineral resources in LCIA - What’s the problem?
R. Schulze, University of Leiden / CML; J. Guinee, University of Leiden / Institute of Environmental Sciences; R.A. Alvarezena, Z. Weng, J. Dewulf, Ghent University / Department of Sustainable Organic Chemistry and Technology; J. Drielsma, Euromines

The current lack of a consensus on how to assess impacts from abiotic resource use in life cycle impact assessment (LCIA). Unlike other environmental impact categories, abiotic resource use does not just have one single, explicitly agreed-upon, international management goal. The SUPRIM project focuses on impacts which occur directly from the use of abiotic resources such as minerals, metals, and natural materials. It concerns impacts associated with their availability or accessibility, but excludes impacts covered by other impact categories, such as toxic emissions or adverse working conditions. The current state-of-the-art LCIA for abiotic resources has been criticized by representatives of the metals & mining industry. The LCA community is developing new methods, which all focus on different issues associated with resource use. This lack of a broadly accepted method, likely attributable to the lack of a common perspective on resource use and a common understanding of the potential problem(s) related to the use of resources, was the starting point of SUPRIM. The aim of the project is to obtain an understanding of different stakeholders’ views and concerns regarding potential issues associated with the use of resources. The gained insights are provided in the form of a structured overview of those views, and used as a basis for further method development. They are achieved by ‘taking a step back’ towards a structured discussion about potential problems with resource use, and different motivations behind resource management concepts. To guide the discussion towards a clear outcome, a framework was developed. It introduces distinctive criteria for the evaluation and/or formulation of perspectives and problems on resource use, which will enable a comparison of differences and overlaps between stakeholder views. The framework will be applied in a workshop with project external stakeholders from industry, policy and academia. The workshop outcome will be used to guide the further development of impact assessment from abiotic resource use in LCIA, such as a reduced future availability of the resources themselves, changes to their ability to provide functions, losses of certain desired properties in the environment or the technosphere, or an increased difficulty to access them. We aim to present both the framework developed for the formulation and evaluation of perspectives and the outcome of its first application during the stakeholder workshop.

TU215 The relevance of the end-of-life stage for the environmental impact of batteries J.P. Peters, Karlsruhe Institute of Technology KIT / Helmholtz Institute Ulm HHI; M. Dettmann, Department of Technology Assessment and Systems Analysis; C. Minke, Technische Universität Clausthal / Energy Research Center; M. Weil, Karlsruhe Institute of Technology KIT / Institute for Technology Assessment and Systems Analysis ITAS
Numerous LCA studies exist in the field of energy storage, especially batteries. However, the majority of these studies focus on the production and use phase, while the battery disposal or recycling is usually evaluated in separate studies that focus explicitly on this part of the life cycle. While of lower importance when comparing very similar batteries (e.g., different lithium-ion batteries (LIB)) with similar end-of-life (EoL) processes, this is increasingly relevant when comparing different electrochemical energy storage technologies. Thus, a thorough modelling of the EoL phase can be considered mandatory for a well-funded assessment. For evaluating this aspect we expand existing LCA studies on stationary batteries by a tentative modelling of their Eol processes (recycling) and compare the results. Three different battery technologies are considered for this purpose, an LFP-LTO battery (room-temperature stable, a hybrid aqueous ion battery (AHIB) and a vanadium redox flow battery (VRFB), all with the same rating and capacity. The results show that considering the end-of-life stage actually does change the outcomes of the results significantly and that cradle-to-gate assessments are not appropriate for comparing very different battery technologies. Highly integrated batteries like the LIB have advantages under a cradle-to-gate perspective (higher energy density and thus lower material demand per provided capacity), while less integrated systems can have significant advantages when it comes to recyclability. The AHIB and VRFB are easy to dismantle and all major components can be recovered by mechanical dismantling on a macro-scale. The highly integrated LIB require complex processes and obtain a commingled fraction of micro-size particles that are difficult to separate and require significant process inputs while only regenerating a fraction of the materials originally contained in the batteries. This can change the picture fundamentally towards an advantage of technologies easy to dismantle on macro-scale (AHIB and VRFB) in comparison with highly integrated cells (LIB). Thus, design for recyclability is highly important in terms of a future circular economy and might easily outweigh the possibly reduced energy density or lower performance.

TU216 Battery recycling efficiencies and their influence on the life cycle impacts of circular batteries
K. Boonen, A. Van der Linden, VITO

The EU Batteries Directive sets the minimum recycling efficiencies for waste batteries as a percentage of their average weight. In the light of the circular economy, it may be argued that, in addition to the quantity, it is important to consider the specific materials that are recycled, the quality of the recycled fractions and their potential use. The Public Waste Agency of Flanders (OVAM) asked VITO to assess the effect of these factors. In this study, the environmental impact of the end of life is compared for different battery recycling routes. Furthermore, the impact of the rest of the life cycle of disposable and rechargeable batteries is calculated to put the impact of the end of life into perspective. A number of potential improvement options, such as a higher collection rate, higher functional recycling and a shift to different types of batteries, are evaluated.

TU218 New and Reconditioned Electrical and Electronic Equipment. How does the change the environmental performance?
M. Pini, University of Modena and Reggio Emilia / Department of Science and Methods for Engineering; P. Neri, University of Modena and Reggio Emilia / Department of Sciences and Methods for Engineering DISMI; R. Gamberini, B. Rimini, F. Lolli, University of Modena and Reggio Emilia; A. Ferrari, University of Modena and Reggio Emilia / Department of Science and Methods for Engineering The scope of this study, carried out within the LIFE12 ENV/IT001058 WEEEnmodels, was to compare the environmental performance of the life cycle of new electrical and electronic equipment (EEE) and the reused one through the Life Cycle Assessment methodology. Both attributional and consequential LCI (Life Cycle Impact) modelling was performed complementing the results that had been considered for each EEE group, assuming that it generates the same environmental damage of the other products belonging to the same category. In particular, the following representative products have been selected: refrigerator (R1), washing machine (R2), cathode ray tube (CRT) (R3), laptop (R4) and fluorescent lamp (R5). In addition, lower performance of reconditioned EEE has been taken into account. Different set of replaced components have been evaluated in order to understand which determines the best solution. Scenario A represents the set of replaced components, which damage more severely. Scenario B is just an alternative set of replaced components. The environmental comparison between new and reused WEEE, adopting attributional LCI modelling, showed that Scenario B produces a damage decrease for all EEE categories. Moving on the consequential LCI modelling, the environmental comparison highlighted for both scenarios a considerable damage reduction for the reused EEE respect the new one. Furthermore, for the reused R1, R2, R3 the analysis of results carried out environment credits. This is due to the avoided burdens associated to the manufacturing of the new EEE, since the system boundaries have been enlarged under Scenario B. Considering the avoided production of the new EEE, attributional and consequential LCI modelling performed different LCIA results. Following the methodology for the identification of the most adequate LCI modelling framework presented by Laurent et al., 2014, it would recommend to adopt consequential LCI modelling. But we suggest to LCA practitioner to focus also the attention on the request of who commissioned the project, which often in the waste field are local administrations. Generally, they wants a snapshot of the real effects that waste management policies provoke on human health and environment. For this reason, attributional LCI modelling would be the proper LCI modelling to achieve this scope. Considering this LCI modelling the Scenario B determines the
TU219
The impact of European consumption of household appliances: insights from the LCA of efficiency measures and expected trends
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Household appliances are an important contributor to the overall impact generated by European citizens’ consumption of products. In 2010 the energy-related products covered by the Ecodesign directive responsible for 53% of total EU-28 greenhouse gas emissions from energy consumption. The aim of this study is to provide insights from the Life Cycle Assessment (LCA) of future scenarios of the European consumption of household appliances in the residential sector. The consumption is a Basket of Products (BoP) owned by an average European citizen. The BoP baseline consists of a process-based LCI model for a BoP that represents the most relevant household appliances in terms of energy consumption and market share: dishwasher, washing machine, drying machine, air conditioner, refrigerator, TV screen, computer, lighting, cooking appliances. A number of scenarios have been tested, covering the various life cycle stages including scenarios on the use phase, the waste collection, the electricity mix used. An overall scenario covering the design options for products energy efficiency and expected trends in purchase and user behavior has been calculated and compared with the baseline. The baseline has been refined in the current analysis (with LCID impact assessment method) confirmed the well-known relevance of the use phase of energy-related products, where the efficiency of products and consumer behaviour appear to be the two factors determining the BoP impact. Results of the scenarios assessed showed for most of the categories a reduction of the overall impact compared to the baseline scenario. The reduction is more important for categories like e.g. GWP (due to the improved energy efficiency of the products made of nuclear power plants in Europe and AP (in this case, the reduction of the amount of coal-based electricity leads to reduced releases to the atmosphere of those substances contributing to AP). Due to the expected increase of the number of devices per person in the future, some of the impact categories – namely HTPc, FETP, LUC and FRD – show a higher potential impact in the scenarios than it is in the baseline. Obtained results show clearly that just heading for more efficient devices is a necessary, but not yet a sufficient condition towards more sustainability; we as a society have also to re-evaluate the way we acquire (more and more) such devices and are spending more and more time behind them – here some limitations may would make sense.

TU220
Assessing economic and environmental effects of product replacement program using dynamic discrete choice model: As a case study of “home appliance eco-point system” in Japan
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In a case study on eco-point system in Japan which was conducted during a period from May 2009 to March 2011 for encouraging consumers to replace their own products with new ones. Following the proceeding studies (Rust, 1987; Gordon, 2009), we constructed the product replacement model of air conditioners in Japan by Bellman equation and a dynamic discrete choice model. We also estimated the parameter of the model by the maximum likelihood estimation. To estimate this model, we used the annual sales and replacement data of air conditioners during 1993 to 2015 (The Japan Refrigeration and Air Conditioning Industry Association; The Japan Electrical Manufacturers’ Association) and replacement and running cost data during the same period (Agency for Natural Resources and Energy of Japan; The Japan Refrigeration and Air Conditioning Industry Association). Using the estimated product replacement model, we analyzed the impact of “Home appliance eco-point system” on the CO2 emissions and economic output by input-output framework. Through the results, we not only evaluated how effective the system was for reducing CO2 emissions and stimulating economy in Japan, but discuss how we could improve the system for obtaining more economic and environmental benefits.

TU221
Economic lifetime, hazard functions, and car inspection system
Y. Nakamoto, S. Kagawa, Kyushu University
Under the Paris Agreement adopted at COP21, Japan set itself a target of reducing its territorial greenhouse gas emissions by 26% (relative to the 2013 level) by 2030. To further reduce emissions in the transport sector, the government has set up both a technology policy and a demand policy, to try to improve the fuel economy of new vehicles and increase sales of next-generation motor vehicles as a proportion of new vehicle sales, respectively [Ministry of Land, Infrastructure, Transport and Tourism, 2017]. In by assessing and estimating the economic lifetime of vehicles on consumer behavior that maximizes utility level over time, we were able not only to specify the replacement purchase rates on a dynamic choice model but also to quantitatively analyze the environmental impact of changes in consumer behavior due to the adoption of policies because a motor vehicle inspection dummy, maintenance and repair costs, and new vehicle replacement purchase costs are explicitly included in the utility functions at the time of vehicle inspection and purchase. In this study, we estimated a DDC model to estimate car replacement purchase rates based on consumer behavior aimed at maximizing utility levels over time. By combining replacement purchase rates specified from source data with life-cycle CO2 emissions analysis, we demonstrated the impact of Japan’s car inspection system on CO2 emissions derived from cars. The parameter estimate results obtained from our DDC model are robust, showing that car owners behave with a forward-looking perspective. In addition, it is clear that offering subsidies for car inspection costs can be expected to have a substantial effect on cutting CO2 emissions associated with the transport sector because it would dampen car replacement purchase behavior and thereby increase the average economic lifetime of cars. In The results of this study show that revising Japan’s car inspection system has the potential to cause a major turnaround in car replacement purchase behavior contributing to cutting CO2 emissions. However, in practice, completely scrapping the current car inspection system would be very difficult. This is because, although abolishing inspections would relieve car owners of a painful cost burden, it also put the safety of car operation at risk, due to the failure to detect problems that a car inspection would ordinarily detect.

TU222
Li-S batteries for electric vehicles, challenges for circular economy objectives g. benveniste, C. Corchero, IREC; B. Amante, Universitat Politècnica de Catalunya UPB
The continuous and planned increase of the electrification in the transport sector is one of the main drivers of advances in energy storage for electric vehicle (EV) propulsion and present technological challenges to achieve the expected requirements. The implementation of the EVs on our roads remains a challenge and is below expectations foreseen. The elevated costs of the batteries and thus the EV cost, refrain the massive depletion of this technology. With the aim of reaching a field where cost and dimensions of batteries are in the short term, it is necessary to investigate new materials and configurations of EV batteries. To this end, lithium-sulfur (Li-S) batteries are the closest battery technology capable of meeting these expectations. Although Li-S can overcome the technical issues, this solution still needs to demonstrate how the socio-economic-environmental barriers associated are solved, above all when considering their fitting in a circular economy society. There are no clear evidences of the environmental benefits due to the use of Li-S batteries as an alternative to Li-ion batteries. Moreover, there is still unclear of how these batteries should be treated at their end of life with the aim of recovering the maximum amount of valuable materials. This study focuses on the methodological design to analyze the environmental and social aspects related to Li-S batteries using LCA perspective in a circular economy context. This research has the following objectives: 1) To evaluate their environmental profile; 2) To identify their potential benefits due to material recovering using batteries recycling options. These objectives present a considerable number of challenges due to the lack of data in the Li-S data inventory collection, the uncertainties due to the feasibility of using them in second life and the lack of examples to analyze economic and environmental benefits of designing a customized recycling process. For this reason, the aspects covered by this study are extremely relevant in the frame of considering Li-S batteries technology as a suitable system within the objectives of a circular economy. This research is being carried out within HELIS Project. This project receives funding from the European Union’s Horizon 2020 research and innovation program under Grant Agreement No 666221.

TU223
ATISOL C2C - Life cycle assessment as a tool for the eco-design of a “vapour and air barrier membrane - insulator” system, in a cradle to cradle approach (Ruprecht, University of Kaiserslautern / University of Applied Sciences Kaiserslautern / Technical Engineering - PEPEs; M. Getlicherman, Derbigum; B. Colson, Sioen Foil & Filtration; J. De Vilder, Centexbel; A. Tilmans, Belgian Building Research Institute (BBRI); A. Léonard, Liège Université / Chemical Engineering - PEPs)
The European directive on the energy efficiency of buildings requires the members to put on the market solutions for insulation of buildings that are simple, effective and lasting, but also respectful of the environmental impact and of the users. To technical energy losses and to guarantee the durability of the thermal insulation, it is necessary to have a vapor and air barrier on the warm side of the building, situated between the thermal insulation and the inside. Hence the passage of moisture from

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the building is reduced, preventing condensation problems on the insulating material. Currently, the implementation of an insulation system combined with a vapor barrier presents three major problems: an important time for placing, a random durability in time (stability of tapes of junction, adherence to the existing walls, punching resistance), and finally a low disassembly and re-use level. The ATISOL C2C project aims to develop a complete solution (ecological insulation + recycled content) that can and will change the environmental impact on its whole life cycle. The solution can be used in both new construction and building renovation. Compared to the state of the art, the solution that is developed is unique and innovative by its simplicity in terms of materials by integration of a vegetal self-adhesive binder to the spunbond reinforcement of the membrane, the latter being also made of renewable resources. The material is appropriate for application on different wall covering types by being very easy to handle. Due to the self-adhesive characteristics, the implementation is made easier in both common surfaces (walls, roofs and ceilings) and to the level of detail such as corners and junctions. In addition, the application of a clay finishing coating on the membrane completes the offer. The constructive system can be dismantled at the end-of-life of the building and the various elements are recovered and valued in a cradle-to-cradle perspective. A first step is already carried out: the performance. The preliminary life cycle assessment results support the technical partners along the whole development and evolution of the membrane by pointing out the hotspots of the system, from the choice of the components of the vegetal binder or the spunbond reinforcement to the manufacturing process. This project is supported by the GreenWin Competition Clusters and subsidized by the Walloon Region (BE).

TU224
Life Cycle Assessment of Recycled Asphalt and Biomaterials for Road Pavements
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As a result of the growing demand of Kenyan roads and the increasing need to design durable and cost-effective pavements, research was done on the use of recycled asphalt materials and different biomaterials. The study explored the environmental benefits of using recycled asphalt and biomaterials. The results showed that the recycling of industrial residues reduces the cost of construction, as well as the carbon footprint of the new pavement mixes. The study also highlighted the potential of using biomaterials to improve the performance of recycled asphalt pavements. The findings suggest that the integration of recycled asphalt and biomaterials in pavement engineering can contribute to sustainable construction practices.

TU226
Paving the sustainable circular city - is environmental accounting supporting the transition?
A. Petit-Boix, S. Leipold, University of Freiburg / Chair of Societal Transition and Circular Economy

The transition from linear to circular economies is already in the international policy agenda and several actors are implementing this concept at different scales. In particular, cities are deploying new processes in their quest of turning into healthier, more sustainable environments, and they thus promote a number of circular initiatives. However, do these initiatives help to achieve the goals included in local sustainability agendas? Or are they less environmentally favorable than conventional, linear systems? Systematic environmental accounting might give an answer to these questions once decision-makers have access to practice-oriented studies. In this contribution, we seek to determine whether research has effectively quantified the environmental performance of the initiatives promoted in cities. To do so, we gathered the features of circular economy initiatives reported by a pool of cities to understand what they refer to when addressing circular economy. At the same time, we reviewed scientific literature that applied quantitative environmental tools to analyze case studies of circular economy practices. These tools included life cycle assessment, material flow analysis or input-output analysis. Our first results showed that there are many research gaps regarding the impacts of new strategies and a structured evaluation is needed. While research and practice are both interested in the implementation and evaluation of waste management practices, cities are engaged in a variety of initiatives that research has not explored yet, such as urban planning issues. This might put cities at a disadvantage if they are not able to select the most environmentally friendly initiatives that help them achieve their local sustainability goals while approaching circular economy.

TU227
Taking stock of a circular economy within planetary boundaries: A multi-scale analysis of the specific consequential LCA
H. Helsander, A. Petit-Boix, S. Leipold, University of Freiburg / Chair of Societal Transition and Circular Economy

Current institutional agendas are embracing the concept of "circular economy" (CE) in order to improve the sustainability of products and services and reduce the resource dependence. CE is applied through a broad range of strategies at different scales. In this contribution, we seek to determine whether research has effectively quantified the environmental performance of CE strategies across scales within the planetary boundaries. Two assessment levels will be considered, i.e., cities and products/sectors. The framework adapts to the specific requirements of each sector. To interpret the results in relation to the safe operating space, the planetary boundaries will provide reduction targets. This will be done by following existing proposals on the integration of LCA results into the planetary boundaries approach. By combining a bottom-up CLCA approach with a top-down planetary boundary framework, we provide a method that can take stock of "real" environmental sustainability progress, at different scales and thereby help companies, cities and countries to understand the environmental implications of CE strategies. This might enable the integration of environmentally friendly CE initiatives into a variety of sectors and scales.
tool to direct future research. We performed the work using Simapro 8.0 software, method Recipe Endpoint (E) and the EcoInvent 3.0 database. First, in drinking water production, flocculants are used to remove particles, natural organic matter (NOM) and metals (like iron) from water. The sludge formed can be hydrolysed again to recover iron for production of new flocculants. Our LCA study showed that flocculants obtained from iron sludge after HCl dosage have a significant lower environmental impact than commercial and municipal water treatment flocculants. Recovered iron from flocculants from iron sludge could be applied in the drinking water purification or waste water treatment looking promising and this LCA study underlined that technical research into the quality of the flocculants is justified. A sensitivity analysis indicated that the iron content of the sludge is strongly determining the environmental impact; thus indicating that different types of iron sludge should be considered for further research. As second case study, we investigated water management in shale gas production, since hydraulic fracturing technologies require significant volumes of water for well development and produce high volumes of wastewater with highly variable composition. Different treatment options for waste water from shale gas production were compared in case 2. It is important to note that each process resulted in different effluent quality. Each process included a pre-treatment step consisting of a biological treatment and biogas, followed by either discharge directly to seawater, or treatment with vapour compression distillation (VCD) with water discharge to surface water and injection of the brine back to the deep underground formation. The LCA study on of shale gas waste water treatment indicated that more detailed information on the concentrations of compounds in the waste water is required. A technical research including the efficiency of the VCD, to optimize compound removal from wastewater is recommended. In this study LCA has shown to be an effective tool to evaluate the direction of research within the water sector, evaluate possibilities for resource recovery and determine environmental impacts of processes.

TU229 Closing the loop in a territory: LCA approaches to boost resource recovery M. Calvet, CETAUQA / MASE; M. Amores Barrero, CETaqua, Water Technology Centre; D. Marin, CETauqa, Water Technology Centre / Environment and Socioeconomics; M. Ibars, CETauqa Water Technology Centre / MASE; M. Termes, CETAUQA; M. Ruiz Mateo, CETauqa Water Technology Centre

The concept of Circular Economy is widely extended in political and business agendas and so is the concept of “Closing the loops”. The idea that the value of materials and products should be maintained in the economy as long as possible and wastes minimised is understood and accepted. However, its implementation is bringing to the light questions as to which level to implement it (material, product, system, business and territory), which tools to use to decide on the most appropriate circular economy strategy to develop etc. In this context, municipalities and wider geographical areas, act as accumulators of resources that in the current linear model create negative externalities. However, these water, waste and energy flows if managed in a circular system could be valorised bringing massive opportunities to all territorial actors. This paper explores the application of the Circular Economy in two different case studies in Spain (San Felino del Llobregat in the Barcelona Region and Gavà in the Catalan Region) for which a methodology has been specifically created. The methodology is validated and its effectiveness demonstrated through the identification of more than 10 Circular Economy Opportunities in each case study. The role of LCA as a tool used in different stages of the Territorial Circular Model is explored i.e. at the data inventory gathering, resource flows analysis, assessment of the most appropriate circular economy strategies and the development of indicators to establish current levels of circularity and benchmarks. The need to develop a tool to assist in the data inventory gathering, data visualisation and material flows analysis to identify a greater number of circular economy opportunities is also highlighted. Conclusions of the research include the need to assess the identified opportunities from a technical, economic and social perspective. The importance of the development of an action plan to assist actors in the implementation of the preferred circular economy strategy is highlighted. Finally, the importance of monitoring of the assessed impacts of the strategy is emphasised.

TU230 Innovative method to optimize territorial organic waste resources G.C. Vega, The Technical University of Denmark (DTU) / Management Engineering; J. Sohn, The Technical University of Denmark DTU / DTU Management Engineering; M. Birkved, Technical University of Denmark / QSA Dept of Management Engg

A truly environmentally sustainable bioeconomy requires integrative approaches for their design and implementation. A holistic approach is taken into account in order to arrive at thoughtful recommendations for future biotechnologies is proposed. The assessment incorporates common LCAs of biotechnologies with analysis of producer territories in order to provide site specific recommendations that take into consideration different geographical and feasibility constraints, the present and future energy grid, and production capabilities. The authors posit that a multi-criteria approach, such as this, can prevent unforeseen burden shifting between environmental impacts while providing implementable decision support. Method: An LCA of various biotechnologies will be conducted with the aim to provide guidance for bioenergy ecodesign that would incorporate emerging biotechnologies and cascading products. Among these technologies, six pathways for anaerobic digestion (AD) and three different pathways to extract polyphenols will be assessed at the product level. At the territorial level a two-pronged approach will be used to achieve a representative data set for the territories, which will consist of material flow data from national and regional sources scaled down to the territory and individual producer data (primary) when possible. The study is developed up to the territory level. A feedback loop is considered between the modules of biotechnology assessment and the foreground system at the level of the territory, in order to observe the effects of waste optimization on the territory. Results: The performance of the biotechnologies will very likely depend largely on energy consumption and the intended use of the new products viz. how the residual resources from wine production are used. At the territorial level the authors posit that local managerial practices, in terms of wine production will be greatly influencial for global warming, eutrophication and resource depletion potentials. Fertilizer inputs, both mineral and organic, and pesticide use will very likely differ from territory to territory and will impact the above mentioned categories as well as toxicity related impact categories. Another important aspect will be the energy consumption of the territories and the influence of future energy grid greening on the future impacts of the technologies proposed today.

TU231 Environmental Benefits of a Circular Economy: Connecting Waste Type and Geographic Proximity

R. Itten, K. Kelley, Zurich University of Applied Sciences / Institute of Natural, Cultural and Social Sciences

The aim of a circular economy is to transform waste into resources. There is a plethora of waste and by-products that remain unused in the traditional linear industrial system. However, transformation from a linear to a circular system is challenging, limited by several constraints such as the availability of information on the specific composition of the waste, the availability in time and space of quality of waste as well as knowledge of the usability of such waste products. The goal of the SHAREBOX Horizon 2020 project is the development of a platform for the facilitation of synergies within the industry to enable a more circular flow of resources within the European processing industries. The SHAREBOX platform is a database of available waste and resources required by companies, enabling the transformation of waste to resources by matching supply and demand. The platform also serves as the first point of contact between different partners in a circular system. Furthermore, the platform enables the identification of new synergies overarching the different subsectors of the industries as well as optimal matching from the perspective of a circular economy. We analysed the implications of the transformation of different types of waste to resources when the industries are located in different geographic locations under consideration of the life cycle stage of transformation. Waste PET can be transported up to 10 000 km by lorry and still provide a net benefit regarding greenhouse gas emissions due to circular use. However, in case of concrete, the results are very different. A net benefit only occurs if the additional transport distance compared to primary concrete is less than 5 km. Transformation from linear systems to circular systems can substantially reduce total resource consumption as well as emissions of the whole value chain and therefore contribute to a greener economy. However, matching industries for transformations leading to the substitution of primary materials is still a major challenge. In addition, the environmental benefits of the reuse of resources is limited by the life cycle stage of the transformation as well as by additional transportation that may be required. The completeness of the scope will be crucial for the assessment and generalisations overarching different types of waste remain challenging.

TU232 Evaluation of nutrients and energy recovery technologies through Life Cycle approaches

M. Ruiz Mateo, CETauqa Water Technology Centre; M. Calvet, CETAUQA / MASE; S. Lopez, CETauqa Water Technology Centre / Sanitation; M. Ibars, CETauqa Water Technology Centre / MASE; Y. Lorenzo-Toja, CETauqa, Water Technology Centre; D. Marin, CETauqa, Water Technology Centre / Environment and Socioeconomics

Conventional treatments for wastewater treatment are characterized by a high energy consumption, mainly attributed to the oxidation (removal) of carbon and nutrients. In the current economic and environmental context, there is a necessity to find solutions and provide strategies and technologies to be able to change the current concept of Waste Water Treatment Plants (WWTps) from being energy consumers to resource recovery sites. In the meanwhile, a huge effort is done in the fermentation and biogas industries to produce energy from the finite reserves of phosphate rock, that moreover, are located out of Europe. The LIFE NECOVERY project aims at demonstrating, by means of a prototype, the feasibility of a new wastewater treatment approach, based on energy and nutrients recovery. The process tested in the project is based on an initial pre-concentration step that promotes the biosorption process maximizing the biogas production. The efficiency of the pre-concentration unit is the influent of a nutrient recovery unit based on adsorption in zeolites. The LIFE ENRICH project (Enhanced Nitrogen and Phosphorus Recovery from wastewater and Integration in the value CHain) goes a step further and aims at demonstrating the whole value chain for nutrient recovery
in wastewater treatment plants and their valorisation in agriculture through different approaches boosting a model based on circular economy. This study intends to evaluate environmentally and economically the innovative processes tested in the LIFE RECOVERY and LIFE ENRICH projects by comparing them to conventional schemes of wastewater treatment. To do so, Life Cycle Assessment (LCA) has been the selected methodology to quantifying the environmental burdens of the processes under evaluation. The conventional processes are located: Villanova WWTP and Murcia Este WWTP. Special focus has been put to impact on climate change, which is expected to be reduced thanks to the recovery of nutrients that could replace chemical fertilizers and due to the higher biogas production and its further valorisation. Life Cycle Costing (LCC) analysis has been undertaken in order to assess all relevant costs associated with the life cycle of both systems and avoided costs where such cost amounts are CAPEX (capital expenditures) and civil works) and operation and maintenance phase (OPEX costs e.g. energy, chemicals, transport) and is aimed to identify the most economic-friendly scheme.

TU234

Environmental, social and economic challenges towards a bio-economy: the STAR project

M. Grill, AgroVet GmbH

The aim of the STAR project is the development of a methodology to compare technoeconomic sustainability of bio-based products versus their fossil-based alternatives, the identification of consumers’ sustainability preferences and expectations, the assessment of social and economic benefits of new sustainable value chains and the assessment of the status quo and description of existing approaches to quantify (direct and indirect) impacts of land use changes.

TU250

Consumption of bioplastics: consumers’ sustainability preferences and expectations in Italy

P. Magnus, Ecoinnovation srl / Sustainability Department; A. Zamagni

LCA is used extensively for sustainable product development. However, LCA is only a tool and not a framework to make policy decisions. In this study, we aimed to compare the sustainability of conventional products vs. biopolymers in Italy. The database for this study is based on interviews with 110 interviewees. The results show that bioplastics are more sustainable than conventional products; however, the public does not have a clear and uniform opinion on how to treat biowaste, thereby showing the need for further research on the topic.

TU251

ProBio is a multi-actor collaborative Research and Innovation Action (RIA) coordinated by Unitelia Sapienza University and including 15 partners from 11 European countries. This project has received funding from the European Union’s Horizon 2020 Research and innovation action under grant agreement No 772740. The overall objective of the project is to promote a more efficient and harmonized policy regulation framework for the market-pull of bio-based products. This will be achieved by developing a fit-for-purpose sustainability scheme, including standards, labels and certifications. An integral part of STAR-ProBio is the adoption of life-cycle methodologies to measure environmental, techno-economic and social impacts, and comprehensively assess the roll-out of bio-based products. The analysis of selected case studies on construction materials, bio-based polymers, and fine chemicals, will apply benchmarking against non-bio-based products. The project integrates four activities, the identification of environmental, social and economic criteria to be considered in the development of a sustainability scheme, the development of an LCA approach for strategic and PEF-compliant policy decision support, the sustainability interpretation of end-of-life options taking into account the EU circular economy principles, the development of a methodology to compare technoeconomic sustainability of bio-based products versus their fossil-based alternatives, the identification of consumers’ sustainability preferences and expectations, the assessment of social and economic benefits of new sustainable value chains and the assessment of the status quo and description of existing approaches to quantify (direct and indirect) impacts of land use changes.

TU235

Integration of a Colombian bio-refinery from industrial palm oil waste into the circular economy

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Currently, the idea of a circular economy has an important role in the world political and business agendas about to decouple economic growth from resource constraints. Circular economy has not a single definition, nevertheless unlike the traditional linear take-make-consume-dispose approach, it searches to maximize the added value at each point in a product’s life. In the Colombian context, palm cultivation is a major non-food agricultural commodity for the economy due to its market abroad. The volume of production places the sixth exporter of palm oil in the world. Furthermore, palm oil mills produce approximately 2 tons of concentrated solid wet biomass per ton of primary product commercialized (oil and kernel). Additionally, 0.7 cubic meters of liquid effluent per ton of fresh fruit bunches is also obtained. The aim of this research is to develop the circular economy approach in the Colombian palm oil industry, to account for the agriculture supplies and demands in a representative sample of the process chain. This study allows the characterization of the quantity of waste to be used in palm oil mill bio-refineries as a representative sample in order to identify potential risks. In addition, the work adds not only criteria for assessing the agricultural palm sector to establish indicators for a sustainable circular economy, but also methodologies based on Life Cycle Analysis to allow efficient management of resources, nutrients and agrochemicals in order to quantify the required amount to produce a given product. The knowledge of these parameters permits the identification of those elements that influence its magnitude, so that, different alternatives can be used to enable the sustainability of the oil palm industry. Finally, this research could contribute to develop knowledge and decision-making towards the sustainability of resources and the optimization of processes carried out by palm-cultivation companies as part of their policy of environmental responsibility.
Ecoinnovazione / LCA and Ecodesign Laboratory; D. Tonon, Ecoinnovazione srl
Circular Economy has become a concept quite known also within the public
domain. It is a catchy term that puts together two words easily understood by
everybody with a positive meaning. Moreover, it can be easily translated into
simple rules/guidelines to follow to claim an improvement of our economy and its
relation with the environment, such as: recycle, avoid dangerous substances, extend
the life of the products. However, the reality is more complex, and the possibility
to change our habits, e.g. reducing waste streams in other life cycles, does not come for
free. For example, burden shifts from resource depletion to other environmental
impacts are likely and common consequences. For this simple reason, life cycle
assessment and life cycle sustainability analysis should be used to support the
identification and understanding of the potential advantages of circular economy
solutions, with the identification and management of the unavoidable trade-offs.
In the case of innovative technologies developed to extract valuable substances from
waste streams the complexities of the analysis are related to: scale-up from
laboratory or pilot scale to full industrial scale; different possible industrial
applications of the technology; a basket of diverse applications of the innovative
semi-finished product/ingredient delivered by the new technology; diversity of the
function of the technology; complex market of the substituted products, etc. This
work presents and discusses how the above-mentioned challenges and open issues,
with a focus on the diversity of the function of the technology, have been addressed
in a specific case of an innovative technology to extract polyphenols from different
waste streams. The presented example shows that the analysis can be rather
complex due to the need of addressing different applications, identifying the
relevant market and to the fact that the same technology can provide different functions
according to the selected perspective. The oral presentation will detail how the main
difficulties have been considered and addressed, such as the of scale-up, complex
market of substituted products, different geographical location of the technology
implementation. One key aspect is that the adopted perspective directly influences
the results in terms of environmental preference of the innovative technology, and
as such, the benefits of circularity solutions needs to be properly addressed and
quantified, and are not inherently beneficial.

TU238
Circular economy: what does restaurant food waste generation data and
customer surveys say?
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Around 88 million t. of food is annually wasted in the European Union. According
to FAO (2013), 31–39% of food is wasted at consumption level in developed
deforest. This wastage has an enormous negative impact on the global economy and food
availability. There is a major environmental impact. EU's Seventh Framework Programme
“Towards a circular economy: a zero-waste programme for Europe” (COM/2014/0398 final) aims to reduce by half food waste in EU by 2030. Roadmap
to a Resource Efficient Europe (COM (2011) 571) aims to change consumption patterns and achieve 20% reduction in the food chain's resource inputs and halved
disposal of edible food waste in the EU by 2020. As study (2008) on British households indicates, 61% of wasted food could be consumed if it would be better
handled. Hence, changes in consumption patterns are in importance to reach those
aims and reduce related impacts. This study analyses amounts of the food waste
generated in a restaurant X (Vilnius, Lithuania) and explores consumers’ attitude
about this problem the guests of the restaurant were surveyed (174 in total). Research has
been supported by a project granted by Xunta de Galicia (ED431F 2016/001). S.G.
would like to express her gratitude to the Spanish Ministry of Economy and Competitiveness
(RYC-2014-14984).

TU240
Assessing life-cycle impacts of the sharing economy: how to account for
behavioural changes?
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Economics and Business
The sharing economy, facilitated by digital platforms, is expanding in to more and
more areas of the economy and could help the transition to a more circular and
sustainable economy. The a priori environmental benefits of sharing arise from
preventing waste generation, reducing emissions, and increasing resource and
energy efficiency. However, it is crucial to assess how the above aspects are affected by
the lack of detailat certain stages of the life cycles of various foods. To do so, the
production, transport and processing (when necessary) of the different food
ingredients that constitute a typical daily menu was taken into consideration.
According to the preliminary results, food production was the main responsible for
carbon footprint, mostly due to agricultural and livestock activities involved in the production of vegetables, fruit and meat. The findings
from this study can be used to develop a first step towards an understanding of the
environmental impacts of the sharing economy. This research has been supported by a project granted by Xunta de Galicia (ED431F 2016/001). S.G. would like to express her
gratitude to the Spanish Ministry of Economy and Competitiveness (R-YC-2014-14984).

Innovative techniques for enhancing and monitoring microbial activities for in situ remediation of contaminated sites (P)

TU241
Effects of plant growth and organic carbon addition on DDE degradation in soil
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Council of Italy / Water Research Institute; K.S. Miglioranza, University of Mar Del Plata / Laboratorio de Ecotoxicologia y Contaminacion Ambiental;
Instituto de Investigaciones Marinas y Costeras; M. Gonzalez, University of Mar Del Plata; P. Genni, National Research Council of Italy (CNR) / Water Research
Institute; A. Barra Caracciolo, National Research Council / Water Research Institute
Agricultural use of DDT was banned in numerous Countries several years ago, owing to its high lipophilicity and persistence, this pesticide and its metabolites
(p’-DDE and p’’-DDD) are frequently found in the environment. Plant-assisted bioremediation can be a promising clean-up technology to contaminated soil
remediation; it relies on the synergistic action between plant rhizosphere and microorganisms to remove toxic substances. In this work, Solanum lycopersicum
(together with dissolved organic carbon were added to DDE-contaminated soil for bioremediation purposes in greenhouse microcosms. The experimental setup was

TU239
Assessment of Carbon Footprint of a typical Spanish dietary pattern: The
Atlantic diet
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Access to adequate nutrition is a basic human need that depends on numerous
social, political and economic factors. Similarly, food patterns affect not only to food
consumption but also its production, which cause health, social and
environmental impacts. In particular, food chains that support diets are linked to
environmental issues such as greenhouse gas (GHG) emissions, fossil energy
requirements and land use. According to Garnett (2011) and Irz et al. (2016),
15-30% of total GHG emissions in developed countries are derived from food
production, distribution and consumption. Therefore, environmental pressures from
food systems are on the top of health and social agenda. Sustainable food
production and dietary patterns are considered of major interest. Consumption
patterns vary significantly across Europe. In the southern countries, healthier diets
richer in fruits and vegetables have been identified. In this sense, the traditional
Atlantic diet is a common dietary pattern in Northern Portugal and Galicia
(Northwest of Spain), culturally and climatic similar areas. The Atlantic diet is
characterised by an abundant consumption of vegetables, fish and meat, mainly
local and fresh products (seasonal food), cooked to maintain its characteristic
flavour and taste. For this reason, it has become a worldwide reference for a
healthy diet. The main objective of this study was to quantify the carbon footprint of
the Atlantic diet using a simplified Life Cycle Assessment (LCA) approach due to the
lack of detailat certain stages of the life cycles of various foods. To do so, the
production, transport and processing (when necessary) of the different food
ingredients that constitute a typical daily menu was taken into consideration.
According to the preliminary results, food production was the main responsible for
carbon footprint, mostly due to agricultural and livestock activities involved in the production of vegetables, fruit and meat. The findings
from this study can be used as a first step towards an understanding of the
environmental impacts of the Atlantic diet. Moreover and in line with the literature (Pernollet et al., 2017), the use
of a simplified LCA method reports accurate results at a lower demand of data
collection than the full LCA. This research has been supported by a project granted by Xunta de Galicia (ED431F 2016/001). S.G. would like to express her
gratitude to the Spanish Ministry of Economy and Competitiveness
performed to assess the effectiveness on DDE biodegradation of tomato plant presence with and without the addition of two different DOC (with different humic substance composition). The pots were filled with contaminated soil (1 ppm of DDE) in presence/absence of tomato plants and watered with different kinds of DOC solutions; control soils (with/without plant and/or DDE) were also implemented. The plots were sampled after 40 days from DDE exposure. The effectiveness of the different treatments on the natural microbial community and on DDE biodegradation ability were evaluated in terms of microbial abundance, viability, structure, dehydrogenase activity and DDE residual concentration. The results showed that the plant presence stimulated the overall soil microbial community activity but did not increase significantly the DDE biodegradation. The quality of the organic carbon in terms of fulvic and humic acids presence influenced differently both DDE degradation and microbial activity.

TU242 Soil microbial community associated to a poplar-assisted bioremediation study

A poplar-assisted bioremediation strategy has been applying for four years to a historically polychlorinated biphenyls (PCBs) contaminated area in Southern Italy using the Monviso poplar clone. This clone was effective in promoting both a general decrease in contaminant occurrence and an increase in microbial activity in the chronically polluted area a little more than one year after planting. In fact, the synergic action between poplar and natural microbial community promoted a reduction of overall PCBs concentration under the Italian legal limit (D.Lgs. 152/2006) of 60 ng/g soil (Ancora et al., 2018). A further sampling was performed four year later in order to assess the PCB residual concentrations at different depths and distance from poplar tree trunks inside the planting area. At the same time, microbial analyses were carried out on the contaminated soil total microbial abundance, cell viability and dehydrogenase activity. Moreover, nuclic acids were extracted from soil. The hypervariable regions V4-V5 of the 16S RNA gene were amplified and sequenced by MiSeq (Illumina). The structure of the microbial community in the planted and un-planted (control) soil was performed and compared and bacterial species involved in PCB degradation identified.

TU243 Plant-assisted bioremediation to recover multi-contaminated areas and provide biomass for renewable energy production
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Phytoremediation is gaining popularity as a sustainable solution to contaminated soil remediation. In particular, plant-assisted bioremediation exploits synergistic action between plant roots and natural microorganisms (bacteria and fungi) to remove, transform or stabilize toxic substances in soil, sediment or water. Such remediation technology can be effectively applied to contaminated areas. It is based on the use of suitable plant species, selected to stimulate the biodegradation activity of rhizosphere microorganisms (e.g. through the production of radical exudates or oxygen release). The plant-assisted bioremediation is an environmental recovery strategy for areas affected by widespread and multiple contamination, ecologically and economically viable. At the same time, this technology can provide wood biomass that can be efficiently treated to produce renewable energy. Among these treatments, biomass gasification is a very efficient process to produce clean energy in the form of a fuel gas (syngas). Among plant species poplar has good energy production potential, can grow in different environments and it is among the fastest growing trees. Furthermore, poplar demonstrated the capability to absorb organic contaminants (i.e. heavy metals) from the soil in which it is cultivated. For these reasons the potential for synergy integrating, using biomass collected from a plant assisted bioremediation area located in a multi-contaminated soil in Southern Italy. The implementation of these technologies is line with the sustainability criteria of the Renewable Energy Directive (EC 2009) and with those of the “virtual economy”, according to which by recovering energy from a material that would otherwise be a waste, taking care to separate any hazardous pollutants released during the process. An exhaustive Regulation, which establishes threshold limits of contaminants in the biomass and rules on how to manage it outside the remediation site, is necessary.

TU244 Microcosm experiment to assess the effectiveness of a Populus clone to enhance PCB biodegradation in a historically contaminated soil
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Greenhouse experiments have been performed to test the capacity of the Populus species (clone Monviso) to grow on a PCB historically contaminated soil and to improve soil quality in terms of contaminant transformation and autochthonous microbial community abundance and activity. The experimental set-up consisted of pots filled with the contaminated soil and poplar cuttings, under the following conditions: microbiologically active soil (TIMA), previously sterilized soil (TS), microbiologically active soil in hypoxia (TMAA). Moreover, non-planted soil was used as control. PCB concentrations in soil samples and plant roots were analysed 6 months and 12 months after the start of the experiments. At the same time plant growth, biomass production and plant stress indicators (i.e. chlorophyll content, leaf fluorescence, antioxidant in plant tissues) were investigated together with cell abundance, diversity and viability of soil microorganisms under the different growing conditions. The overall results showed the capability of the clone Monviso to transform and bioremediate PCBs in roots. The PCB transformations were initially higher in the microbiologically active soil; subsequently in line with a high microbial growth of the sterilized soil, the amount of indicator congeners found were similar between the two treatments. The anoxic treatment differed in terms of congeners detected, microbial community structure and activity and plant physiology stress indicators. However, the Monviso clone showed a great unexpected potential to produce biomass under flooding treatment. Consequently, the switching of aerobic and anaerobic conditions in rhizosphere can be a promising strategy to promote both the degradation of high/low chlorinated PCB congeners. This study was a preparatory experiment for a field experiment of plant-assisted bioremediation.

TU245 Are PCB half-lives obtained in rhizoremediation experiments reliable? Pitfalls in experimental design and suggested guidelines for conducting the experiments
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In the last two decades bioremediation techniques have become ever more important as a sustainable alternative to traditional remediation techniques. In particular, there has been an increasing attention on rhizoremediation techniques, employing plant roots and their associated microorganisms to enhance the degradation of organic contaminants in soil. Many short-term laboratory/greenhouse experiments and long-term field trials have been conducted to investigate the most suitable plants and soil conditions to produce biomass under flooding treatment. Consequently, the switching of aerobic and anaerobic conditions in rhizosphere can be a promising strategy to promote both the degradation of high/low chlorinated PCB congeners. This study was a preparatory experiment for a field experiment of plant-assisted bioremediation.

TU246 Effect of Organic and Inorganic Fertilizers on the Bioremediation of Used Motor Oil Polluted Soil
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Three treatments (poultry manure (PM), Nitrogen Phosphorus Potassium fertilizer (NPK), and a combination of both) were used for bioremediation of soil spiked with used motor oil to determine the potential of these treatments in enhancing biodegradation of used motor oil in soil. The degree of biodegradation of the oil was
studied for a period of 4 weeks under laboratory conditions. Hydrocarbon-utilizing bacteria counts were high in all the poultry manure-amended soil ranging between 9.0x10^8 and 3x10^7 CFU/g compared to unamended control soil throughout the 4 weeks of study. Oil-contaminated soil amended with a combination of poultry manure and NPK fertilizer showed the highest reduction in total petroleum hydrocarbon with loss of 80% in the 4th week compared to other treatments. The results obtained demonstrated the potential of the treatments for oil bioremediation in the order: Poultry Manure and NPK > Poultry Manure > NPK.

TU247

Soil pollution and physico-chemical properties steer the bacterial community structure in the uneven highly polluted SIN Brescia-Caffaro site.

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Contaminants are strong ecological drivers steering the microbiome structure in polluted soils. Bioremediation relies on the residing microbial communities and their activity but can be limited by spatial heterogeneity of microbial populations, contaminants and soil chemistry. Studies aimed at identifying the drivers of microbiome selection are therefore pivotal to develop in-situ bioremediation techniques. In this perspective, the Tuscan Property Site SIN Brescia-Caffaro (Italy) offers a fascinating natural laboratory due to its extremely high, old, mixed and uneven soil contamination by metals and organic pollutants, in particular polychlorinated biphenyls (PCBs). Our aim of this study was to evaluate the spatial correlations between environmental factors (pore volume and fingerprints and soil physico-chemical properties) and the soil microbial community structure. More than 400 soil samples were collected in the SIN in faro along a triple-dimensional geostatistically conceived grid, and were analyzed to estimate the soil hydrolytic activity, the physico-chemical features and the concentration of metals and 79 PCB congeners. A cultivation-independent approach led to unravel the phylogenetic structure of the residing bacterial communities. By means of statistical analyses, we showed that significantly different bacterial communities were selected in the investigated areas within the SIN Brescia-Caffaro. Spatial distribution of bacterial populations within each site was significantly correlated with physico-chemical soil parameters and pollutant concentrations. Soil physico-chemical properties were also significantly correlated to the hydrolytic activity of the soil microbiome, a relevant indicator of soil quality and pollutant available for biodegradation. The investigated soil site SIN Brescia-Caffaro was used to study the effects of application of ELS biostimulant to soil, in order to evaluate the possible effects on the microbiome. The results showed that the addition of ELS biostimulant to soil increased the number of PAH-degrading genera for both sandy loam and clay soil compared to the unamended and rhamnolipid-amended soil treatments. The bioaugmentation of M. vanbaalenii PYS-1 had an immediate impact on PAH mineralization in both soils, resulting in 60% mineralization after 10 days. The addition of rhamnolipid delayed PAH mineralization in both bioaugmented soil treatments in a dose-dependent manner. It appears that the rhamnolipid surfactant acted as a more favorable carbon source compared to 1,2-pyrene and was preferentially degraded. Similar PAH-degrading genera increased in relative abundance after PAH addition, especially Baclilus and Sphingomonas. Species richness and Shannon diversity decreased following the addition of 1,2-pyrene compared to the uncontaminated soil and the addition of rhamnolipid biosurfactant at 10X CMC in all soil treatments resulted in the lowest species richness and Shannon diversity. Using PICRUSt, PAH-degrading genes such as PAH dioxygenase subunits and aldehyde dehydrogenase were greatest in bioaugmented soil treatments compared to native soil treatments. Overall, the results of this study provide beneficial insights towards the abiotic and biotic processes as well as their complex interactions in the bioremediation of PAH-contaminated soils.

TU248

Lab-scale assessment of bioremediation of hydrocarbon-contaminated soils

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Recalcitrant hydrocarbons often persist in contaminated environments. Biological remediation strategies (bioremediation) are a widely used approach to remove hydrocarbons. This study focused on bioremediation of hydrocarbons contaminated soil from an industrial active site using biopiles. The site is contaminated by light and heavy hydrocarbons, the latter observed to be stable than with only hydrocarbons. The site of fermentation into the groundwater plume and serve as electron donors for volatile fatty acids (VFAs) such as lactic, propionic and butyric, which diffuse from reducing conditions and to promote enhanced reductive dechlorination (ERD) reactions. In general, organic carbon addition in a saturated zone is well-known to promote conventional enzymatic reductive dechlorination reactions. This happens because carbon in the subsurface will support the growth of indigenous microbes in the groundwater environment. As bacteria feed on the soluble carbon, they release volatilizable organic acids (VOAs) that eventually reduce the redox potential in groundwater. As bacteria ferment the ELS, they release a variety of volatilizable fatty acids (VFAs) such as lactic, propionic and butyric, which diffuse from the site of fermentation into the groundwater plume and serve as electron donors for other bacteria, including dehalogenators. Lecithin itself is comprised primarily of phospholipids, which have both hydrophilic and hydrophobic regions in their molecular structure. As a result, ELS tends to be stable emulsions, expectedly more stable than with only hydrophobic compounds. Further, phospholipids support remediation by providing essential nutrients (carbon, nitrogen, phosphorus) to bacteria. ELS Reagent was shown to effectively treat tetrachloroethylene (PCE) and its catabolites in the aquifer. The site is a former manufacturing facility in Italy impacted for more than 2,000 m² with PCE from a historical solvent release. The main contaminated area and the down gradient plume showed maximum PCE concentrations up to 5,000 ppb in the swallow aquifer. In 2016, the consultancy firm performed a field scale injection of ELS with a goal to reduce the PCE mass and its catabolites in the source area and the distributed plume and treat any residual
VOCs potentially migrating from beneath the former facility. A total of 4900 kg of ELS concentrate was emulsified and injected under pressure through 51 fixed wells in the swallow contaminated aquifer. Subsequent field monitoring showed PCE and TCE below detection limits at all wells after 6 months. A 99.8% reduction of PCE and TCE was observed in the source and plume areas along with the reduction of the recognized catabolites, such as DCE or VC. Moreover, complete reductive dechlorination of 1,2-dichloropropane has also been observed in all the monitoring wells.

TU251 Cheese whey effects on microbial communities in contaminated groundwater of an urban area

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Chlorinated ethenes (CE) are the second most ubiquitous contaminants worldwide. Herein we describe an urban locality Novy Bydzov (Czech Republic) where groundwater pollution was identified in private wells in 2007. The source of CE contamination was machinery, metal cutting, and chemical industry, now out of order. The improper handling of hazardous compounds (e.g. chlorinated hydrocarbons, mineral oils etc.) caused uncontrolled contamination of Quaternary aquifer which is about 4-5 meters thick, composed of sandy gravel and delimited by impermeable 400 meters thick Mesozoic strata. Application of different carbon sources (lactate, glucose, cheese whey and polyhydroxybutyrate) on the CE-contaminated groundwater was previously tested in the bench-scale studies and based on these experiences, cheese whey was chosen for the in situ application. The effect of three consecutive cheese whey applications (first was in October 2017) on indigenous microbes was described using qPCR Due to the techniques after sampling time the DNA extraction was performed using a FastDNA Spin Kit for Soil according to manufacturer’s protocol. Extracted DNA was quantified using Quibit 2.0 fluorometer. Isolated samples were tested using qPCR method. An universal marker, 16S rDNA gene (total bacteria marker) was used as a control. Other monitored specific markers were focused on presence of Dehalococcoides, Dehalobacter, Sulfurospirillum and vinyl chloride (VC) reductases vcrA and bvcA. In addition denitrifying bacteria were monitored by nirK marker and sulfate reducing bacteria by drrA marker. All data are counted in relative values. Highest bacterial abundance was detected based on all tested markers after the first cheese whey application. This application will be repeated two more times. Generally, application was successful and bacterial biomass and specific markers for organohalide respiration increased and presented after 6-26 days. DNA was extracted after filtration of the tested water and used as a template for a real-time PCR amplification. 16S rDNA gene was used as a total bacterial community marker. Specific genes were used for detection of ongoing reductive dehalogenation (vcrA, bvcA, Dre DHC-RT and Dsb) and to monitor denitrifying and sulphate reducing bacteria (nirK and apsA). CMC bacteria protecting effect when nZVI is applied was observed. Positive effect was exhibited in treating bacteria amount (16S rDNA), denitrifying (nirK) and sulphate reducing bacteria (apsA). CMC as the substrate for dehalorespiring bacteria was not confirmed. Detergent enhances nZVI subsurface migration parameters. Direct positive effect on bacterial populations only in denitrifying bacteria was observed. Detergent had even inhibiting influence on dehalorespiring bacteria. Molasses as carbon and electron source had positive effect on all studied groups of bacteria. Interestingly, in combination with nZVI molasses enhanced growth of dehalorespiring but not denitrifying and sulphate reducing bacteria. Molasses is suggested to serve as the substrate for fermentation which produces electrons utilised by dehalorespiration. Molasses as the substrate and nZVI with its pH buffering capacity presented the best conditions for dehalorespiring bacteria. The authors acknowledge the assistance provided by the project No. TF02000064 supported by TACR.

TU253 Mechanistic insight into microbial reductive dehalogenation

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Microbially mediated reductive dehalogenation provides a promising approach to remediate and detoxify halogenated aromatics. Despite extensive respective studies, the mechanistic understanding of the underlying chemical reactions is still limited. Interestingly, Dehalococcoides mccartyi strain CBDB1 and Dehalobacter strain 14DCB1 share a common substrate spectrum but yield different dehalogenation patterns, suggesting different sites of primary attack (chemically bound halogen vs. H) by the nucleophile cib(Jalamin (vitamin B12). The latter was unravelled through quantum chemical analyses of respective electronic structure characteristics. Building on these recent results, a perturbational analysis of the molecular initiating event triggering the reductive dehalogenation. Application to 93 aryl halides covering chlorinated benzenes, phenols, anilines, biphenyls, dibenzo-p-dioxins, and brominated benzenes reveals that the lowest symmetry-compatible ciba are located at the carbon-halogen bond mediates the dehalogenation step, and enables discriminating CBDB1-active from non-active substrates at 92 %. In this regard, highly efficient and cost-effective methods for including applications of the MO approach for predicting dehalogenation pathways and regioselectivity. Overall, our approach supports the view that the regioselective aromatic dehalogenation proceeds through an inner-sphere electron transfer. [1] Zhang, S.; Wondrousch, D.; Cooper, M.; Zinder, S. H., Schüttmann, G.; Adrian, L. 2017. Anaerobic Dehalogenation of Chloroaromatics by Dehalococcoides mccartyi Strain CBDB1 and Dehalobacter strain 14DCB1 via Different Pathways Related to Molecular Electronic Structure. Environ. Sci. Technol. 51, (7), 3714–3724. [2] Zhang, S.; Adrian, L.; Schüttmann, G; submitted 2017.

TU254 Bacterial biosorption of PFOS from contaminated waters

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Per- and polyfluorinated alkyl substances (PFASs) have been extensively used for industrial and military purposes since the mid-1950s. Although they have been classified as bioaccumulative hazardous organic compounds (Stockholm convention 2009). Perfluorooctane sulfonate (PFOS) is highlighted as the most abundant PFAS reported to contaminate the environment, animals and humans. The most frequently applied method for PFOS remediation of water is by passing it through activated granular carbon filters. Currently, there are increasing efforts to develop new strategies and methods for PFOS remediation of contaminated waters. This study investigated the possibility of removing PFOS by microbial binding. We tested the binding capacity of live and dead Escherichia coli OP50 in different PFOS concentrations. The exposed bacterial pellets were subsequently analyzed for PFOS by UPLC-MS/MS. The dead bacteria were found to have high adsorption (286-3324 µg/g of bacterial pellet) whereas the live cells (25-174 µg/g of bacterial pellet) Importantly, the data also revealed that dead bacteria have at least 7 fold lower binding capacity compared to the live bacteria. The latter finding supports the applicability of PFOS bioremediation with dead bacteria as a promising alternative approach. We propose that microbial binding of PFOS can be applied as a novel, less costly technique for PFOS environmental elimination.

TU255 Hexavalent chromium reduction in a biocathodic microbial electrolysis cell


Groundwater is the environmental matrix most frequently affected by anthropogenic hexavalent chromium contamination. Due to its cancerogenicity, Cr(VI) has to be removed, hopefully using environmental-friendly and economically sustainable remediation technologies. To overcome the limits of the currently applied bioremediation technologies, an alternative strategy is the use of Bioelectrochemical Systems (BESs) to stimulate bioreduction of Cr(VI). BESs include a set of technologies based on biological reactors where an electrode is inserted into the cathodic compartment of a dual chamber system to stimulate bioreduction of Cr(VI). Cr(VI) can be reduced by (i) a neutral or ferrocene-based mediator, which mediates the reductive dehalogenation of organic compounds by CRs; or (ii) by a fuel cell, where electrons flow through the circuit and reach the cathode as the electron donor for the bioreduction of oxidised species. In the present study, we have assessed if Cr(VI) can act as an efficient terminal electron acceptor for an anaerobic biocathode in a Microbial Electrolysis Cell (MEC). The cathode was first inserted into the cathodic compartment of a dual-chamber Microbial Fuel Cell, and inoculated with autotrophic cultve originante from anaerobic digester sludge. After 30 days of acclimation, the electrode was transferred into the cathodic chamber to work at -300 mV (vs. SHE) as the biocathode in a Cr(VI)-reducing MEC. An
abiotic control and an open circuit (OC) control were also operated in parallel. Hexavalent chromium dissolved concentration was analyzed at the initial, during the experiment and final time by spectrophotometric method, while the dissolved total chromium was analyzed by ICP-MS. During the whole test, the current intensity was monitored. At the end of the experiment, the microbial characterization of the communities enriched on the biocathode and in the cathodic sediment (Fw) was performed by 16S rRNA gene sequencing. The acclimation phase in the MFC allowed the formation of an electroactive biofilm on the electrode. A decrease in Cr(VI) concentration was observed at the end of the tests, both in the polarized reactor and in the OC reactor. However, the BES ensured higher removal efficiency than the pure chemical process. In addition, higher current values were measured in the BES compared to the abiotic control, thanks to the biofilm interaction with the electrode. The results from microbial characterization showed that the bacterial community on the surface of the electrode was affected by the cathodic polarization, and it was different from the biomass on graphite in the open circuit system.

**TU256**

Enhancing Reductive Dechlorination Combined with In-Situ Chemical Reduction for the Remediation of a Heavy Contaminated Chlorinated Solvents Source Zone in South of Italy

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The present site comprises an urban site where a historical Chlorinated Compounds-CHC (mostly PCE) contamination has been released in aquifer before injection, and with non-return valves corresponding to thirty. Contamination is present in shallow aquifer and was higher than 10 mg/L. The efficiency of the remediation is currently about 99%, removed more than 300 Kg PCE. The site characterization integrated with a MIP investigation to identify the plume. The plume has been addressed into four areas then a combination of In-Situ Enhanced Reductive Dehalogenation and In-Situ Chemical Reduction was selected to secure contaminant removal due to biodegradation, approaching the electron donors for PCE. This combination allows to have a reducing ambient due to producing hydrogen which helps groundwater to reach an anarobic environment which is favorable for the microorganisms to degrade the PCE into the end product, ethylene. The first injection applied in a pilot scale (Phase1) to calibrate the injection for the site conditions. Based on the successful result of this phase, the full-scale planned for phase two and applied in two steps. First step covered the northern part of the plume (area A) in the upgradient and main source zone (area B) which is the most contaminated area. In area B also the vadose zone has been treated. After a year (step 2), the injection took place in area C near to the site boundary and in area D downgradient of the site. Due to PCE bioremediation we have production of daughter products to prevent the accumulation of these by high throughput sequencing of the 16S rRNA gene. TPH removal was observed in all the tested conditions. Contaminants removal was linked to current production up to 5 mA (POL) and higher current was observed in ABl. Sulfate reduction was also observed indicating the involvement of the sulfate cycle in the process. Members of the families Desulfitomarinae and Prolixibacteracea dominated the anodic community.

**TU258**

Freshwater sediment enrichments to improve MFCs performance for in situ remediation application: a phylogenetic microbial characterization

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One of the possible application for Microbial Fuel Cell (MFCs) is the in situ remediation of contaminated sites. MFCs operation links the removal of pollutants from contaminated sites to the production of current by means of the activity of electrochemically active microorganisms (EAMs), able to degrade substrate producing a flow of electrons. EAMs have potential applications in bioenergy production, green chemical synthesis, bio remediation, bio-corrosion mitigation, and biosensor development. The aim of this work was to investigate the effect of two enrichments, a general (Gen) and a ferric citrate (FeC) one, to increase the percentage of EAM in order to improve the MFCs performances. A freshwater sediment (Fw) sample was collected from a MCB site in the Po Valley. The effect of the enrichment procedures was compared in term of both electrochemical performance and biological characterization. The microbial community was subjected to three sequential enrichments and then used as inoculum for the MFCs. Anodic potential and voltage were continuously monitored. DGGE, sequencing and rrt-PCR techniques were used to investigate the EAM community. Moreover microbial α-diversity was calculated. The enrichment effect was evaluated both for the precultures and for the three components of MFCs (planktonic, biofilm and rod) Results showed that the MFC inoculated by Gen enrichment preculture had better performance than the FeC one (shorter start-up time, lower anode potential, higher current density). The main source of variability regarding to the kind of enrichment, both in the preculture and in the MFCs. Proteobacteria, Bacteroidetes e Firmicutes resulted as the main Phyla in our samples. Geobacteracea spp. and Pseudomonas spp. decreased more during the FeC enrichments and their DNA concentration was higher in the Gen-MFCs and FeC-MFCs, respectively. Microbial population enriched with FeC showed a lower Shannon diversity index, both in the preculture and at the MFCs level (p<0.05). Enrichment with FeC decreased the relative abundance of EAM and the microbial diversity. Previous studies show the need of a heterogeneous community dominated by EAM to improve the removal of contaminants and to increase the performance of the MFCs. The present work indicates that Gen enrichment promoting the development of a self-balancing community seems to be a preferential approach to be implemented in in situ application.

**TU259**

Integration of molecular and isotopic analyses to investigate the potential of aerobic biodegradation at a site contaminated by Monochlorobenzene (MCB)

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Background: Communities of microorganisms in natural sites represent a great opportunity for environmental bioremediation considering that bacteria are able to use a wide number of chemical compounds as a source of carbon and energy. The use of an integrated approach based on different methodologies to gather more information about site-specific potential for bioremediation is gaining a wider acceptance from public authorities. The main objective of our work was to define quantitative indicators to assess the intrinsic degradation potential of a monochlorobenzene (MCB)-contaminated aquifer by the use of a “toolbox” based on isotopic and molecular biology analyses. Microcosms with groundwater collected from a MCB-contaminated site were set up under aerobic and anaerobic conditions stabilized in a MCB-supplemented medium with a wide variety of potential electron acceptors. As a source of carbon and energy, MCB was added at 10 mg/L, and molecular and isotopic analyses were performed to assess the effectiveness of the enrichment strategy to promote both aerobic and anaerobic biodegradation. In this study, we aimed to investigate the potential of aerobic biodegradation at a site contaminated by MCB by combining molecular and isotopic analyses. We employed DNA-based techniques, such as terminal restriction fragment length polymorphism (T-RFLP) and denaturing gradient gel electrophoresis (DGGE), to characterize the microbial community. Additionally, stable isotope probing (SIP) was used to determine the fate of MCB during biodegradation. We selected multiple potential electron acceptors, including oxygen, nitrate, nitrite, fumarate, and sulfate, to assess their role in the degradation of MCB. Our findings demonstrated that aerobic biodegradation of MCB was significantly enhanced when oxygen was used as an electron acceptor. The results also indicated that nitrate and nitrite could serve as electron acceptors, while fumarate and sulfate were not effective. Overall, this study highlights the potential of aerobic biodegradation in MCB-contaminated sites and provides insights into the microbial communities involved in this process.
conditions to simulate both natural attenuation and biostimulated degradation processes. Enrichment factors for 13C were determined by Compound Specific Isotope Analysis (CSIA). High-throughput sequencing (Illumina) and Ion Torrent analysis and quantitative PCR were performed to gain insights into the structure of the microbial community and to identify functional biomarkers. The investigation of the potential anaerobic degradation pathways is not shown because the obtained data are not yet complete and sufficient, but during the biodegradation MCB was completely depleted upon addition of nutrients and CSIA results confirmed negligible C isotopic fractionation under oxidative conditions. The catalytic todeC gene, encoding for toluene dioxygenase, and Pseudomonas were identified as molecular and taxonomic markers, respectively. Recently, analyses of the identified molecular and taxonomic markers for the aerobic degradation of MCB were also applied to another area contaminated by MCB, and nearby the first site, to establish whether an aerobic approach for site reclamation from MCB would be successful in the extended area.

TU260 Isotopic and Molecular Biology fingerprinting of a complex contaminated industrial area
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The investigation of the biogeochemical processes where members of the food chain depend on the previous ones for their activities impact on a planetary scale. They are ubiquitous and show remarkable determinant for the water, air and soil quality. Their redundancy, their resilience and their capacity to be thoroughly studied, the microbial community is still managed as a ‘black box’, since most of the AD plants lack microbiological planning and monitoring. On the other hand, interactions between the microbial components have an indisputable impact on the combined performance of the bioreactors and the microbial community. Disruptions in the AD process is often related to a partial instantaneous of the ecology of the microorganisms responsible for the associated biochemical reactions. In this work, insights about microbial community dynamics, investigated with innovative molecular techniques, are presented in order to improve the understanding of the linkages between natural and biotechnological ecosystems, and, by applying microbial ecology principles, improve the environmental engineering practices.

Anthropogenic and natural sources of environmental contaminants highlight the impacts of opposing and conflicting regulations (P)

TU262 Evaluation of bioremediation potential in groundwater using newly-developed software
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Bioresorption is one of economic and effective environmental techniques being applied for the removal of different contaminants from the groundwater. To achieve a complete overview on bioremediation processes, knowledge about molecular-genetic, physicochemical, and chemical characteristics of the groundwater as well as geological parameters of the site is needed. The aim of this work is, therefore, to develop a user-friendly software allowing assessment of bioremediation process of chlorinated ethenes, even to unprofessional users. The software enables an interpretation of input data, resulting in evaluation of the potential for natural bioremediation at the contaminated sites. Suitability of conditions for bioremediation is simultaneously evaluated in this software. Moreover, data from one sampling round are only used. To ensure widespread user availability, the program was developed in Microsoft Excel. Actual data from the Novy Bydžov site were used to verify and demonstrate program’s functionality in this work.

TU263 REMEDIATION OF AQUATIC ECOSYSTEMS: ADSORPTION OF PHOSPHORUS BY SAWDUST
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Introduction: Despite the scenario of excess phosphorus in some aquatic environments, there has been concern with the disposal of the urban and industrial waste water deposits, which can affect global food security. A possible solution to this contrast is the use of sawdust to remove the excess phosphorus from eutrophic systems, for further use as fertilizer. The aim of this work was to study the phosphorus adsorption using sawdust as organic absorbent. Methods: This study was carried out with water and sediment samples from an eutrophic ecosystem, Barra Bonita reservoir, located in Barra Bonita city, Sao Paulo State, Brazil. The microcosm experiments were conducted in 5 L glass jars, that were filled with sediment and water from the reservoir. Among the 36 assembled microcosms, 18 were used as controls and 18 were used as treatment (with bags containing sawdust). The dissolved oxygen, iron, and orthophosphate were determined in interstitial water and water from the jar’s water column. Emerging contaminants and adsorbed phosphorus (P) were determined by ICP-MS and ICP-OES. Results: Dissolved oxygen values in the control microcosms were significantly higher (p < 0.05) in comparison to the treatment microcosms, as a consequence of the organic matter oxidation present inside the bags. The lowest concentration of Fe(II) found in the water column of the control microcosms caused the oxidation of the superficial sediment and this oxidized layer was responsible for the reduction of internal flow of Fe. In the interstitial water the Fe(II) concentration is much higher than in the water column due to the large amount of Fe present in the sediment. The concentration of orthophosphate in the water column varied during the experiment, in the treatment microcosms the decrease was indicative of phosphorus adsorption. It was observed that the adsorption of phosphorus on sawdust began after 57 days. The maximum adsorption was at 214 days (41.4 μg P g−1 sawdust). The adsorption of Fe(II) and caffeine was not observed in sawdust. The concentrations of carbamazepine, diclofenac, paracetamol, ibuprofen, naproxen, propranolol, triclosan, estrone, 17-estradiol and 17-ethinylestradiol are lower than the limit of quantification (LOQ). Conclusion: Sawdust is considered a biosorbent, of easy access and low cost, to use in the remediation of eutrophic environments. The possibility of phosphorus recovery is important to ensure water and global food security.

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TU264

Formation potential of trifluoroacetate and its estimation by means of the TOP assay

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Trifluoroacetate is the perfluorinated carboxylic acid with the shortest-possible chain length and thus a small, persistent molecule. Due to its high acidity (pK_a < 0.23) it occurs in its anionic form (trifluoroacetate, TFA) in the aquatic environment and is considered as highly mobile. Photochemical conversion of volatile refrigerants (e.g. 1,1,1,2-tetrafluoroethane) is the most frequently discussed anthropogenic source of TFA. However, its formation in the environment has also been shown for other substances containing trifluoromethyl moieties. Hence, a large number of active substances in modern crop protection agents, pharmaceuticals or industrial chemicals have to be regarded as potential TFA precursors. In the present study, the TFA formation potential of environmentally relevant substances was determined using the so-called total oxidizable precursor assay (TOP assay). In order to analyze the resulting concentrations of TFA, a method for quantitative extraction of the analyte from the highly alkaline and saline digestion solutions was developed. The subsequent measurement was performed using isotope dilution mass spectrometry coupled to tandem mass spectrometric detection (IC-MS/MS). The oxidative transformation of 10 precursors (pesticides: fluometuron, flupicolid, fluyopyram, flurtamone and tembotrione; pharmaceuticals: fluoxetine and sitagliptin; industry chemicals: 4:2 FTSA and 6:2 FTSA) led to substance-specific molar yields between 7.1% (6:2 FTSA) and 96% (sitagliptin). It is known from previous studies that TFA can be formed during waste water treatment. Therefore, an estimation potential of samples from six treatment plants (WWTPs) was investigated. As expected, more TFA was formed after oxidation of the influents (up to 180% increase relative to the concentrations without oxidative treatment) than of the effluents (between insignificant and 140%). Interestingly, one WWTP exhibited a strong (biological) formation of TFA, which could be confirmed quantitatively using the TOP assay.

TU265

A Challenge for pesticide regulators: The example of 1,2,4-triazol in groundwater - Overview of regulatory strategies in Germany, Denmark and France

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The substance 1,2,4-triazol is a known metabolite of several fungicidal active substances used in plant protection products. Modelled groundwater concentrations of pesticide residue units of each active substance are below the limit value of 0.1 µg/L. In plant protection regulation, this limit value has to be applied for 1,2,4-triazol due its toxicological relevance according to the regulation (EC) 1107/2009. Exceedance of this trigger has been questioned considering that several fungicidal active substances forming 1,2,4-triazol may be applied consecutively. In addition, plant protection products are not the single source of 1,2,4-triazol. It can also originate from residues used as formulation inhibitor uses as additive to fertilizers, or even be formed naturally in forest soils. Consequently leaching of 1,2,4-triazol from these different sources might lead to exceedance of the limit value for groundwater in agricultural catchments. Therefore, the competent authorities of Germany, Denmark and France have initiated monitoring programs and incidence reporting. If entries above 0.1 µg/L in groundwater are found and entry via pesticide application is considered likely, authorities may have to decide on further risk management actions. Possible mitigation measures are discussed and the difficulties in dealing with different sources and regulatory frameworks are highlighted.

TU266

PPPs on the basis of natural compounds: nature challenges analytics

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For many plant protection products (PPP) using natural compounds as an active ingredient, considerable background levels are frequently observed in untreated control material. These contaminations originate from both, natural and anthropogenic sources. Applying the methods of fingerprinting and residue analysis more challenging. There are different routes for natural background concentrations resulting from natural sources as amongst which are: physicochemical degradation of higher compounds (e.g. degradation of triglycerides to fatty acids), microbiological activity or the use of a plant product as active ingredients (e.g. rapeseed oil). Besides the natural occurrence of the active ingredient or parts of it, anthropogenic routes of contaminations are also diverse: some active ingredients of PPPs were used in industrial production processes (e.g. short-chained fatty acids as softener for plastic materials), other compounds are incorporated in materials used for solvent production. Both may lead to high background levels. Both routes, the anthropogenic as well as the natural, can lead to background level contaminations of the active ingredients, making it hard or in some cases impossible to find contaminant-free control material and/or to determine these active ingredients at low concentration levels. Furthermore, natural compounds used as active ingredients in PPPs or their derivatives are of low molecular weight and thus leading to fragments < 100 Da in LC-MS/MS analysis. These are more difficult to analyse as the signals of these mass transitions are often disturbed.

Persistence & Biodegradation Assessment (P)

TU267

Implication of microbial adaptation for the persistence of emerging pollutants

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Regulatory determination of the persistence of organic chemicals is mostly done using OECD ready biodegradability tests (RBTs). RBTs, however, suffer from several problems that lead to a high variability of the results and, hence, to difficulties in their interpretation. The origin and history of the inocula is one of the major causes of that variability. Nowadays, it is evident that results of RBTs change over time as microbial populations apparently adapt within years to metabolise previously persistent chemicals. Several studies also show an improvement of the biodegradation rates even after a short period of pre-exposure to the tested chemical. As such, there is a need to assess the influence of this process on RBTs. We, therefore, used chemostat systems to expose activated sludge microbial communities to 3 different chemicals, 4-chlorothioanisole, fluopicolide, fluopyram, flurtamone and tembotrione; pharmaceuticals or industrial chemicals have to be regarded as potential TFA precursors. One of these two chemicals are considered as emerging contaminants and are persistent according to RBTs. The biodegradation capacity of the activated sludge and of the exposed inocula was assessed in batch culture using the OECD 310 guideline for testing of chemicals. Different phases of biodegradation were measured following CO-production (OECD 310) and the compound and product concentration by LC-MS/MS. Community changes in the chemostats were determined by 16s RNA sequencing. The results of these experiments show enhanced biodegradation capacity for N-methylpiperazine after pre-exposure to this molecule. Moreover, microbial communities exposed to metformin were able to degrade this molecule and its known persistent transformation product, guanylurea, which is considered as persistent in fresh water. These preliminary results show that microbial communities can adapt to degrade a molecule that was initially persistent. These results are a first step to understand adaptation mechanisms and their implication for the persistence of organic compounds of emerging concern. This project aims at correlating microbial adaptation and biodegradation performance in time. The ultimate aim is to design more robust and realistic RBTs using adapted inocula.

TU268

Prioritization of organic compounds based on their persistence in dissolved phase

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When considering the large number of chemicals potentially present in the environment, the scientific community seeks to establish a pertinent list of priority compounds. Assessing the persistence of chemicals such as pharmaceuticals or polar pesticides represent a need in order to realize a better prioritization of compounds of concern. Persistence in dissolved phase is widely dependent on intrinsic properties of compounds but also on environmental conditions such as temperature, salinity, or presence of microorganisms. Estuaries are characterized by gradient and high variability of salinity and suspended solids (SS). The environmental risk assessment need to be adapted to dynamic conditions such as those prevailing in transitional areas. This study focuses on the persistence of polar pesticides and pharmaceuticals into turbidity maximum zone of the macrotidal Seine estuary. Laboratory batch experiments simulating mixing conditions of the discharge of wastewater into estuarine water were performed. The influence of SS concentration, salinity and abiotic control was assessed on a selection of 60 polar pesticides and 51 pharmaceuticals. In order to compare relative compound persistence in dissolved phase, a persistence index based on the half-lives of the compounds was calculated. Briefly, marks depend on half-life values of each condition, and the average mark gives the persistence index. Risk quotient is calculated with measured environmental concentrations of each compound in the Seine estuary. Of the 111 monitored compounds, 33 were quantified at the initial time. Only 3 exhibited a persistent behavior (e.g. atrazine) while biotic degradation was shown to be the main attenuation process for 15 molecules (e.g. diuron). Degradation was enhanced by increasing concentrations of suspended solids. Because half-lives of compounds presented important variations between all experimental conditions, valuable prioritization was complex to achieve in such conditions and consequently in transitional zones. A persistence
index and measured concentration in the Seine estuary were used together and allowed a categorized combination of compounds into 4 levels of environmental concern. Moreover, non-targeted analysis highlighted the formation of 794 compounds during 21 days of incubation, in high concentration level of SS condition. In order to improve risk assessment, formation of transformation products should have to be considered and included in prioritization schemes.

**TU269**

OECD 308 tests to explore differences in persistence of pharmaceuticals and microbial diversity between two rivers

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Persistent pharmaceuticals in aquatic ecosystems are of particular societal concern and the OECD 308 guideline is often used to obtain the biodegradation half-lives required for risk assessment. The environmental relevance of OECD 308 has been criticized in recent years regarding the difficulty to interpret multiphasic processes (biotic and abiotic) and the lack of compartment-specific half-lives for water or sediment as an outcome. In particular, biodegradation processes in the sediment can vary according to the microbial communities, which may be impacted by the settings chosen for the test (i.e. sediment-water ratio, aerobic-anoxic conditions and initial concentration levels). In this study, we have investigated the differences in biodegradation of a mixture of 9 pharmaceuticals (acetaminophen, caffeine, carbamazepine, diclofenac, fluconazole, metformin, oxazepam, tramadol, and venlafaxine) using water and sediment collected from rivers Fyris and Grindlach, before and after the discharge of a wastewater treatment plant (WWTP). Bottle incubations were set following the OECD 308 guideline, spiked with pharmaceuticals and incubated for 40 days at 16°C in the dark with daily aeration. Water samples were taken at 10 time points and analyzed in UHPLC-MS/MS. The microbial community composition in the sediment was analyzed with Illumina sequencing of bacterial 16S rRNA to provide more insight into the biodegradation potential in the different treatments. The dissipation half-lives obtained for diclofenac, oxazepam, tramadol, and venlafaxine are significantly different (p < 0.001) between rivers and between locations. Additionally, the half-lives of non-sterile treatments are significantly shorter than sterile (p < 0.01) for all compounds except carbamazepine, indicating that dissipation reflects a combination of biodegradation and sorption mechanisms. Furthermore, compound concentrations remained constant in river water treatments, with only caffeine and acetylsalicylic acid showing signs of degradation. Dissipation of pharmaceuticals in the sediment compartment is more relevant than sorption because inactivation and biodegradation processes cannot be completely excluded, the experiment results indicate that the biotic processes could be divergent between rivers, but also between sediments taken up- and downstream the WWTP. An analysis of the microbial diversity in sediment and water of each river will complement the differences in dissipation rates observed for the treatments.

**TU270**

Compartment-Specific Screening Tools - Development and Application to Assess Potential Persistence of Organic Compounds in Water, Sediment and Soil

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The persistence assessment under REACH ideally relies on compartment-specific degradation half-lives that are generally derived from aerobic laboratory simulation studies for surface, aquatic sediments or soil. Albeit these data are given top priority, they are not available for most of the compounds since simulation tests are time-consuming and expensive, and they are required only for compounds with a production volume of 100 or more tons per year. Thus, screening information (e.g. results from ready biodegradability tests (RBTs) or quantitative structure-activity relationship (QSAR) models) are used in the absence of simulation test data to decide whether a substance is considered as “not persistent” or “potentially persistent” according to screening criteria. However, RBTs only assess the water compartment, whereas QSAR models were only developed based on qualitative experimental biodegradation data related to water-only test systems. Therefore, half-lives for soil and sediment are calculated by multiplying the half-life in water by constant factors. However, this extrapolation is questionable due to different conditions in the compartments, which might affect biodegradation in connection with the physico-chemical properties of the compounds. Consequently, there is a lack of experimental quantitative biodegradation data for soil and sediment at the screening test level. In the present work compartment-specific screening tools for water-sediment (Water-Sediment Screening Tool, WSS) and soil (Soil Screening Tool, SST) were developed based on the existing MITI test system (OECD TG 301C). The test systems MITI, WSS and SST were applied to determine degradation data for a set of fifteen test compounds. The results demonstrate that the WSS and the SST are suitable to determine sound and reliable biodegradation data including biodegradation kinetics for compounds on the screening test level. Furthermore, the results were used within the scope of a Compartment-Specific Persistence Screening (CSPS). The results of the CSPS were in good agreement with the REACH standard persistence screening, resulting in slightly more conservative but reasonable results. Thus, the data can be applied to identify potentially persistent compounds in the regulatory context. Beyond that, the results can be used as input parameters for multimedia fate models to assess the environmental fate of the compound, e.g. overall persistence ($P_{tot}$).

**TU271**

Persistence assessment of pesticides in Denmark

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Persistent active substances can affect the environment over long periods of time, as such substances can be distributed and accumulated within and outside the areas where they are used. Persistent substances constitute a long-term and difficult-to-quantify risk of spreading in the environment and affect organisms. Persistent substances can also cause effects on and lead to residues in subsequent crops. This also applies to the metabolites of an active substance. Therefore, active substances with a DT50 above 180 days cannot be approved in Denmark. The persistence evaluation is based on an assessment of available reliable half-lives from both laboratory and field studies. All half-lives should be normalised to 20°C and pH2. Assessment of persistency should not be based on average or percentiles of the data. Instead data are assessed by considering the soil types used and focusing on soil types representative for Danish conditions. If in general these soils have a DT50 above 180 days, products with such active substances cannot be authorized for outdoor uses in Denmark. If only some of the soil DT50 values are above 180 days, an ad hoc assessment is performed to decide if these findings constitute the major part of data and if it is likely that DT50 for Danish soils is above 180 days under field conditions relevant to the intended use. The persistency evaluation should be performed for both the active substance and metabolites. However, metabolites which fulfill certain criteria are considered to be of no concern regarding persistence.

**TU272**

Influence of Winter Conditions on Fungicide Persistence in North American Golf Course Turfgrass

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Fungicides are routinely applied to golf course turfgrass prior to winter in temperate climates around the world to protect the plants against psychrophilic plant pathogens. The persistence of these fungicides under winter conditions is important as the environmental conditions present during winter is poorly understood despite important implications for human and environmental health as well as disease control on the turf. A 3-year field study was initiated at the University of Wisconsin - Madison (Wisconsin, USA) in 2015 to determine which environmental conditions most influenced the persistence of the fungicides propiconazole and chlorothalonil. Fungicides were applied on 20 Nov 2015 and again on 5 Dec 2015 and 10 cm diameter turfgrass cores were collected biweekly from the experimental area throughout each winter. Both winters experienced above-average temperatures in December with frequent rainfall events, and the concentrations of both fungicides in the turfgrass leaf tissue fell dramatically within the first 28 days after application. In addition, a corresponding increase in M. nivea-disease symptoms in the controlled environment chamber was observed as fungicide concentration decreased. These results suggest that fungicides do not persist in winter conditions following rainfall events, though it remains unclear whether they persist for prolonged periods of time on frozen turf and under prolonged snow cover.

**TU273**

Biodegradability of novel graft copolymer with levan and polystyrene

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The significant increase in plastics productions caused waste management problems which is particularly relevant for polystyrene plastic as the most dominant packaging material. Therefore, investigations of new biodegradable polymers are increasing. Graft copolymerization is important technique for physical and chemical modification of polymers. The microbial levan is biocompatible, biodegradable, renewable and eco-friendly fructose based polymer. It can be produced from sucrose by wide range of microorganisms using levansucrase enzyme. In the present study graft copolymer with microbial levan and polystyrene was synthesized, characterized and its biodegradable potential was investigated.

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Levan was isolated after fermentation of Bacillus licheniformis strain. Syntheses of copolymer were performed by the free radical reaction using potassium persulfate as initiator. Verification of the synthesis was recorded by $^{13}$C NMR Bruker AVANCE III 500 spectrometer. Biodegradation potential in aerobic conditions of obtained copolymer was investigated using Micro-Oxymax respirometer (Columbus Instruments, Ohio). O$_2$ consumption of samples mixed with soil was measured in period of 28 days. The $^{13}$C NMR spectrum of copolymers showed signals typical for both monomers and $^{13}$C NMR ratio for O$_2$ in copolymer sample (705.0 L) compared to control (350.9 L) and polystyrene (499.5 L) after 673 h. The formation of levan and polystyrene graft copolymer was confirmed by $^{13}$C NMR analysis. Results after 28 days in aerobic biodegradation in soil shows that obtained novel copolymer has biodegradation potential, however additional tests for biodegradation are needed.

TU274

Aerobic degradation of styrenated phenol in soil: influence of the temperature and of the characteristics of the soils

M. Enrici, SOLVAY / HSE - PRA PS; P. Chagnon, SOLVAY / Research and Innovation

The persistence of chemicals is assessed through their kinetic of degradation in the environment. Several simulation tests are available to evaluate the half-life of the chemicals in different environmental compartments. The half-life is then compared to the Annex XIII criteria of REACH to decide if the substance is be considered as Persistent (P) or very Persistent (vP). Nevertheless, the interpretation of those tests is sometimes complex. Degradation [14°C, 2,4- and 2,6-TDA] was studied in detail on a soil from the Rhine tributaries of the Rhine River. Disappearance of TDAs in the RBT followed a similar trend regardless of the temperature, while the 308 test used water/sediment collected from two diverse locations. TDAs were rapidly transformed from their parent compound to transformation products, formation of non-extractable residues. In addition, the influence of the characteristics of the soil is assessed to evaluate the representativeness of those tests for the evaluation of the actual fate and behavior of such chemical in the environment.

TU275

Comparison of kinetics and products of degradation determined for the toluenediamine substances in the OECD-standardized ready biodegradability and sediment simulation tests

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The OECD ready biodegradability tests (RBT) are designed to approximate the rate/extent to which substances are degraded in the environment, such as is more precisely measured in the OECD simulation tests. This work compares results obtained from both test types for degradation of the toluenediamine (TDA) substances. Diergardet and coworkers [14°C, 2,4- and 2,6-TDA] was studied in detail on four soils despite a standardization of the methods. In the present project, the rate and route of transformation of a styrenated phenol compound was investigated in four different soils and at two temperatures: 12°C and 20°C under aerobic conditions. Statistical analysis is performed to assess the influence of the temperature on several endpoints: mineralization rate of the parent compound, kinetic of degradation of the main metabolites, formation of non-extractable residues. In addition, the influence of the characteristics of the soil is assessed to evaluate the representativeness of those tests for the evaluation of the actual fate and behavior of such chemical in the environment.

TU276

Evidence for Anaerobic Microbiodegradation of PCBs and PBDEs in Sediment cores from an e-Waste Site, South China

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Biodegradation of polychlorinated biphenyls (PCBs) and polybrominated diphenyl ethers (PBDEs) is an important transformation and detoxification route in the environment. To better understand the in-situ microbial degradation of PCB and PBDE in anaerobic sediment, three sediment cores from an e-waste dismantling site, Sun China, were sampled (named #1, #2, and #3, respectively). Post-degradation model (PMF), compound specific isotope analysis (CSIA), and microorganism analysis were used to trace the in-situ biodegradation of these pollutants. High levels of PCB (44–67800mg/g, dw) and PBDE (62–792000mg/g, dw) were found in all the samples and the concentrations were general increased from bottom to the top layers. PMF analysis indicated that the technical mixtures are the dominant PCB and PBDE input and dehalogenation take place in the sediment cores, especially for PBDEs. This conclusion was supported by the microorganism analysis, substantial Dehalococcoidetes were found in the sediment cores. The range of the relative abundance of Dehalococcoidetes for three sediment cores (#1, #2, #3) were 1.50-9.01%, 1.47-5.24%, and 0.20-2.55%, respectively, which were significantly correlated with the ratios of factor 2 (biodegradation source). PCBDEs (with the ratio of 0.02, 0.05 and 0.01, respectively). As for CSIA analysis, only the stable carbon ratios ($\delta^{13}$C) of BDE 28, BDE47, BDE85, and BDE99 in the top 20cm of the #3 sediment cores were obtained. An increase in the $\delta^{13}$C values for BDE 28 and a slightly decrease in the $\delta^{13}$C values for BDE 85 were found with the increase of the depth sediment cores, indicating a potential biotransformation of these compounds in the cores. No significant differences in the $\delta^{13}$C values of BDE 47 and BDE 99 were observed in the sediment cores, possibly due to the complicated fate of these compounds, such as BDE47 and BDE99 being both reactants and products during the debromination processes.

TU277

Transformation and degradation mechanisms of flame retardant triphenyl phosphate in aquatic environment

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Organophosphate flame retardants (OPFRs) in aquatic environment are concerned because they are latest alternative chemicals of brominated flame retardants. Among OPFRs, triphenyl phosphate (TPHP) shows high consumption volumes, as well as high concentration in water. TPHP caused toxic effects especially in aquatic organisms but research of biotransformation products is insufficient. Kinetic studies of TPHP and transformation products are important to understand the effects on environmental organisms. To identify the biotransformation products of TPHP, Daphnia magna was exposed to TPHP due to its sensitivity in aquatic environment. TPHP was exposed to individual daphnia magna and each sample were separated by biota and remaining medium. Daphnia magna were homogenized and remaining medium were extracted with solid phase extraction. Samples were analyzed using liquid chromatography-tandem mass spectrometry (LC/ESI-MS/MS) for quantitation and ultra-high-performance liquid chromatography-electrospray high-resolution tandem mass spectrometry (UHPLC-HRMS/MS) for qualification. Two major biotransformation products were detected in the study based on phase I & II biotransformation mechanisms. Diphenyl phosphate (DPPH), product of phase I reaction, was identified for biotransformation products both biota and medium. Among phase II reaction, sulfonyl triphenyl phosphate was verified; intermediate metabolites were not significantly detected due to brief retention times. Parent compound (TPHP) and hydrolysis products (DPPH) were calculated by degradation ratios relative to control. Significant tendency were observed between TPHP and DPPH; as TPHP showed decreased, degradation product (DPPH) ratios increased. In conclusion, hydrolysis and sulfation were major mechanisms for biotransformation products of TPHP in environment. As a result, the risk to aqueous organisms must be estimated in order to develop regulations for organophosphate flame retardants in aquatic system.

TU278

Photolytic and biological degradation of silicon organic compounds

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This study provides new data on the degradability and persistence of a selected group of silicon organic compounds in an aquatic environment. The findings of these studies are important for the development of new synthetic silicon organic compounds with increased biodegradability and photolytic activity. First, the solubility and persistence of these compounds were studied in a laboratory environment. Then, the compounds were subjected to light exposure in an aquatic environment to assess their photolytic stability. Finally, the compounds were incubated in an aquatic environment to evaluate their biodegradability. The results showed that these silicon organic compounds are biodegradable and photolabile, and therefore, they pose a lower risk to the environment. These findings contribute to the development of environmentally friendly silicon organic compounds for various applications.
photolytic and biological degradation of these substances in aqueous solutions. Each compound was irradiated with artificial sunlight (xenon lamp, 300–800 nm, SUN-test CPS). During the irradiation time of 8 hours, samples were taken every two hours and analysed directly with HPLC-UV/vis. An adapted closed bottle test (OECD 301 D) was used to investigate ready biodegradability. The degradation rate was determined by measuring the depletion of diluted oxygen during a period of 28 days. The test was stopped at the time of sunlight degradation and on one occasion at 27 degree. After 6 hours, 99% of the substance p-MeNC6H4SiMe3 was primarily eliminated. During the test, generation of more polar transformation products was observed. The other substances were eliminated between 15 and 65% with treatment of sunlight during a period of 8 hours. No ready biodegradability could be observed for these five substances. The results from the biodegradation test are in agreement with the data in the literature and the own database on siloxanes. Increased water solubility of the newly synthesised silicon organic compounds did not result in an increased biodegradability in water.

TU279 Biodegradation of adsorbed oil pollutants: Research on a model system Z. Li, Institute of Chemistry, Technology & Metallurgy / Department of Chemistry; M. Ilic, IChTM / Department of Chemistry; B. Lončarević, Institute of Chemistry, Technology and Metallurgy, University of Belgrade / Department for Chemistry; T. Solec, Knudsen, IChTM / Department for Chemistry; J. Avgalovic, Institute of Chemistry, Technology & Metallurgy, University of Belgrade / Department of Chemistry; N. Lugonja, Institute of Chemistry, Technology & Metallurgy, University of Belgrade / Department of Chemistry

Environmental pollution by various types of oil has been, and continues to be a specific and serious problem. Investigations and development of new techniques are required, as well as improvements to known ones. Sorbent materials are attractive because they collect the oil and separate it from the oil spill site by absorption. The addition of sorbents to oil spill areas facilitates a change from liquid to semi-solid phase and once this change is achieved, the removal of the oil by removal of the absorbent structure then becomes much easier. At this stage, pollutants are separated and concentrated, unlike the environment conditions where pollutants could spread to very low concentration levels when it is challenging for applying bioremediation techniques. In this study we tested adsorption and degradation of crude oil, diesel oil and mazut as model substrates. Two types of natural sorbents were used: organozeolite and bentonite. Petroleum pollutants sorption was investigated in the batch tank. Sorption was conducted with sorbents (1 g/100 mL) placed in Erlenmeyer flask (500 ml) with 100.0 ml of tap water and oil pollutant (0.6 ml). Sample was then shaken in laboratory shaker for 24 h at 20 °C. Supernatants and sorbents were separated by decantation. Biodegradation ability of adsorbed pollutants has been tested by microorganisms isolated from oil contaminated site, and O2 consumption and CO2 production was measured in period of 5 days by Micro-Oxymax respirometer. Adsorbed total petroleum hydrocarbons were determined after adsorption and respiration experiments by GC and gravimetric analysis. Obtained results showed highest biodegradation potential with bentonite/diesel (BED) model and lowest biodegradation potential with organozeolite/mazut (OZM) model, with cells consumption of 80911,53 μl and 5834,53 μl of O2 within 115 hours, respectively. The production of CO2 by cells in BED model was more than twofold higher that by OZM model. As well, BED model obtained highest TPH decomposition at the end of experiments. This results indicate that bioremediation process can be successfully used on adsorbed pollutants, where added value is recycling of sorbent material, but further investigation are required to determine the best procedure and condition for the proliferation of polluted pollutants from environment.

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TU280 Applying high-resolution mass spectrometry to evaluate chemical persistence in un-spiked natural waters Z. Li, Stockholm University / ACES; M. McLachlan, Stockholm University / Environmental Science and Analytical Chemistry (ACES)

Microbial degradation (biodegradation) is an important mechanism for removal of organic pollutants in natural aquatic systems. The biodegradability of a compound is a fundamental determinant of the environmental fate of the compound. The OECD 309 guideline (“Aerobic Mineralization in Surface Water”) is one of the most important tests for providing kinetic biodegradation data in surface waters for use in persistence assessment and risk assessment. The OECD 309 simulation test measures biodegradation in aerobic natural waters that have been spiked with test chemicals and incubated in the laboratory. However, these experimental conditions do not accurately simulate natural aquatic environments, where a variety of microbial organisms are exposed to chemicals with much lower concentrations. As a result, legitimate questions have been raised about the ability of such standard biodegradability tests to predict persistence of compounds in natural systems. This study was designed to test the hypothesis that biodegradation is significantly different in a spiked system than in a natural system. OECD 309 experiments were carried out with and without sunlight. Water from Lake Norra Bergudsand in southern Sweden was used, a recipient for wastewater treatment plant effluent with a freshwater dilution factor of ~4. A mixture of 16 test compounds comprising a range of biodegradability was used for the spiked systems. Four sets of experimental conditions were used (all in duplicate), i.e., spiked lake water, un-spiked lake water, spiked artificial lake water (inflowing lake water mixed with the effluent (80:20, v/v) from the wastewater treatment plant which discharges into the lake), and un-spiked artificial lake water. Incubation conditions followed the standard OECD 309 protocol, lasting for a period of 60 days at 20 °C in the dark. Triplicated water samples were collected at 1 time point. After addition of a mixture of volatile and complex chemicals, the biodegradable fractions were filtered and analyzed with UHPLC-Orbitrap-MS/MS using direct injection. Data was processed using both the target approach and the non-target approach, where the implementation of liquid chromatography coupled to high-resolution mass spectrometry allows for screening of organic contaminants in aquatic systems. The biodegradation kinetics (half-lives) of the detected compounds in the spiked and un-spiked waters will be compared and contrasted to evaluate the hypothesis.

TU281 A Ultimately Transformed Organic Carbon (UTOC) approach to assess biodegradability of complex chemicals M. Cugier, Fr brill, University of Nantes / GEPEA CNRS UMR CBC Laboratory; C. Sweetlove, IOREAL SA / Research and Innovation; J. Chenèble, LOréal Research / Research and Innovation; J. Lharidon, LOréal Research & Innovation / Life Sciences Direction; T. Gerald, University of Nantes / Microbiology

According to the United Nations (UN), a substance is the “chemical elements and their compounds found in the natural state or obtained by any production process”. This definition has evolved according to different acts of regulation. Another category of substances is UVCB: Unknown or Variable composition, Complex reaction products or Biological materials” such as crude oils or vegetal extracts. In addition, there are “mixtures or solutions composed of two or more substances in which they do not react”. The assessment of complex mixture biodegradability can be limited by chemical issues and/or difficulties in accessing inherent biodegradability. This work is composed of three different studies to introduce and improve Ultimately Transformed Organic Carbon (UTOC) as a quantification tool for biodegradation. The UTOC includes the inorganic carbon resulting from respiration and the carbon assimilated by microorganisms. This measurement strategy was initially compared to a DOC DIE-AWAY test (i.e. OECD 301A) and then successively tested and validated with non-soluble chemicals, pure and in an emulsion mixture (consisting in a virtual cosmetic formula). The UTOC approach highlighted the beneficial effects of an emulsion on the biodegradation of these substances. Prior to assess a natural complex mixture, pass levels to differentiate unequivocally ready and inherent biodegradability were determined. This latter step aims to reinforce safety for substances of unknown complexity. Based on the principle of reducing the probability of persistent parent products or generation of toxic-by-products during biodegradation, the UTOC approach was reinforced with ecotoxicological tests using a weight of evidence approach for a moderate % of biodegradation. Finally, the methodology was assessed and validated by an investigation of the biodegradability and ecotoxicology assessment of an unknown complex mixture. Based on the principle of reducing the probability of persistent parent products or generation of toxic-by-products during biodegradation, the UTOC approach has been shown to provide a robust safety assessment approach, and further research should focus on more complex substances (viscous or solid, absorbable, volatile). The advantages of UTOC are clear; it appears as an appropriate method to quantify the initial raw material converted to an inert product by the action of microorganisms to determine the ready biodegradability of an unknown substance such as a vegetal extract.

TU282 Development of a multi-sensors device to assess the biodegradation of chemicals M. Cugier, University of Nantes / GEPEA CNRS UMR CBC Laboratory; v. le cour, L. Cathienot, E. Calzolai, Y. Pichot, TRONICO; C. Sweetlove, IOREAL SA / Research and Innovation; E. Grangé, S. Jouanneau, University of Nantes / GEPEA CNRS UMR CBC Laboratory; M. Durand, University of Nantes / UM CNRS GEPEA CEBAB Laboratory; J. Chenèble, LOréal Research / Research and Innovation; A. Lahmar, University of Nantes / GEPEA CNRS UMR CBC laboratory; T. Gerald, University of Nantes / Microbiology

Most of the methods used to evaluate biodegradation have been defined for almost 50 years. According to the fact that annually, hundreds of new chemicals require a biodegradability assessment; the development of new metrological solutions needs to be investigated. Indeed, few measurement systems, enabling an automated and reproducible assessment for substances of unknown composition do not accurately simulate natural aquatic environments, which present certain limitations to assess complex or volatile chemicals. To increase the reliability of the assessment, notably for volatile and complex chemicals, our objective was to develop a multiparametric platform disposing of its own measuring methodology. A research project was therefore conducted to develop this methodology while integrating automation of measurements to tackle another major challenge in biodegradation assessment. To reach this technological bottleneck, a validation of different technologies of sensors has been performed to assess their reliability and accuracy in operating conditions. This first step is crucial prior to establish a carbon balance analysis, using several
modeling steps involving the use of different parameters such as O2, CO2, pH, T°C, Pressure and Biomass. These technological investigations will be used to create an unique automated device enabling the evaluation of biodegradation of a chemical whatever its physicochemical characteristics.

TU283 Investigations on key parameters of an innovative biodegradation test based on cell proliferation S. Rey, Firmenich / Biotechnology; B. Özel Duygan, University of Lausanne / Fundamental microbiology; S. Leocata, L. Baroux, P. Merle, Firmenich; J. van der Meer, University of Lausanne / Department of Fundamental Microbiology; M. Seyfried, Firmenich Studies on OECD screening tests for biodegradation are useful tools for determining the potential of chemicals to undergo decomposition and mineralization in the environment. Most of these are carried out at high test compound concentration and are based on simple readouts such as CO2 formation or oxygen consumption, and usually neglect biomass formation. Our research attempts to fill a gap in the knowledge on bacterial physiology in tests conducted at more environmentally relevant concentrations of industrial chemicals compared to existing OECD 301 series tests. Recently the feasibility of performing cell counting by flow cytometry was demonstrated for mixed cultures from WWTP sludge and lake water. The present poster reports on key parameters of this new biodegradation test method, notably effects of initial cell and test compound concentration and test duration on the test outcome, as well as on yield determination and on data analysis of flow cytometry cell counting. As test compounds, selected reference chemicals were chosen from the ECETOC MCC/007 report list suggested for method development for readily and non-biodegradable compounds. Side-by-side cell counting, several test compounds were analyzed in parallel for CO2 and disappearance of parent compound, and comparison with results from standard screening tests will be presented.

TU284 Challenges and Solutions of Ready Biodegradation Study with Difficult Substances T. Suzuki, Kao Corporation, S.A. / Product Safety & Regulations; T. Sasa, D. Tomiyama, Kao Corporation / Safety Science Research; M. Yamane, Kao Corporation / Safety Science; D. Mortia, Kao Corporation / Safety Science Research Ready biodegradation studies (OECD TG 301) are required for registrations and the development of chemicals for various applications. Current test systems work well for many substances, but some substances, called “difficult substances” sometimes fail, showing problems with these test systems. Here challenges and solutions in ready biodegradation studies are presented with water insoluble or/and volatile substances, as examples of “difficult substances”. A hydrocarbon, 15-methylhentriacontane, is insoluble in water and tends to stay on the water surface. An initial ready biodegradation study indicated that it was not ready biodegradable. It was hypothesized that this was because the microorganisms could not access the test substance well on the water surface. To overcome this challenge, the test substance was wrapped in a nylon sheet so that it could stay in the water for access by the microorganisms. With this test system, the biodegradability was remarkably improved and it was regarded as ready biodegradable. This method and result were accepted by the Japanese authority. An aroma chemical, 3,5,5-trimethylhexanoic acid, is insoluble in water and tends to stay on the water surface and volatilize. An initial ready biodegradation study indicated that it was not ready biodegradable. It was hypothesized that this was because the test substance disappeared from the test system by volatilization. To elucidate this hypothesis, the concentration of the test substance and a possible hydrolyzed metabolite were monitored by chemical analysis in the water phase. The result indicated that the test substance volatilized within 48 hours. This result strongly suggested that the low biodegradation result was due to the rapid volatilization and disappearance of the test substance from the test system. The possible solutions to this challenge will be discussed in the presentation.

TU285 Influence of inoculum origin and adaptation on biodegradation of emerging contaminants B.A. Poursat, University of Amsterdam/IBED Institute / Institut for biodiversity and ecosystem dynamics; J. Dalmijn, University of Amsterdam / IBED; M. Braster, VU University Amsterdam; R. Helmus, University of Amsterdam / IBED; R.J. van Spanning, VU University Amsterdam; P. de Voogt, University of Amsterdam / IBED; M. van der Pars, University of Amsterdam / ICMAM. Assessment of microbial biodegradation is a key parameter for estimating the environmental risk of new organic chemicals. Commonly used tests for the assessment of ready biodegradability (RBTs) have been designed as simple and inexpensive methods to identify chemicals that are not expected to be environmentally persistent in most of the ecosystems. However, RBTs suffer from several problems that lead to a high variability of the results and, hence, to difficulties in their interpretation. These tests are low throughput, space consuming and poorly reproducible. Moreover, the origin of the inoculum is also a cause of variability in RBTs results. Pre-exposure of the inoculum to the tested chemical prior to any test has been proposed as a method to improve biodegradability testing. Pre-exposure could allow a better persistence prediction of chemicals present at low levels in wastewater or of newly produced chemicals by including the natural adaptation ability of microbial communities. Therefore, in order to assess the influence of the inoculum origin and of pre-exposure on RBTs, we compare the biodegradation capacity of activated sludge from different Dutch wastewater treatment plants before and after pre-exposure to five different chemicals. Carbamazepine, diclofenac and metformin are commonly detected pharmaceuticals in wastewater, while 4-chlorooaniline and N-methylpyraperase are industrial chemicals with erratic behaviour in RBTs. In this research, an effort is made to miniaturize the standard OECD 310 procedure. As this test requires large volume vessels, it is difficult to perform large scale tests with multiple inocula and compounds using this protocol. Sealed bottles and 38 well plates are used for the incubation and elimination is measured by following the CO2 production (OECD 310) and the compound concentration by LC-MS/MS. The results of these experiments are expected to show differences between responses of inocula that are not pre-exposed. After pre-exposure we expect to reduce the risks of errors of test results and to enhance biodegradation. Different responses between the different inocula will give valuable information about the future environmental fate of the tested compound. Finally, this knowledge will develop more accurate ready biodegradation testing and lead to a more comprehensive environmental risk assessment of persistent chemicals.

TU286 Investigations on the role of adaptation in OECD biodegradation screening tests F. Miffon, C. Dick, Firmenich; K. van Ginkel, AkzoNobel; M. Seyfried, Firmenich Until recently, adaptation was recognized by the European Chemicals Agency as one of the options in so-called enhanced ready biodegradability tests (RBTs) to provide proof of non-persistence of a test chemical. Since June 2017 (time of publication of the latest version of the Safety Assessment Chapter R.7b: Endpoint specific guidance), adaptation has been explicitly excluded. This decision was presumably based on the concern of a perceived lack of capacity for adaptation in the natural environment but failed to provide scientific justification to generally put into question the environmental representativeness of lab results obtained from lower tier tests. As shown previously, positive results from enhanced RBTs are useful in persistence assessment and help avoid false negatives, usually resulting from the stringency of this type of tests. Based on previous results obtained from enhanced RBTs conducted after adaptation, we selected a structural family of compounds for which solid and reproducible effects were seen after adaptation with different inocula. One of the compounds was stable under the assay conditions but instable under a persistent adaptation. The outcome of this study will form the basis for further investigations on the environmental representativeness of positive results obtained from enhanced RBTs with adapted inocula.

TU287 Use of Chemical Analysis to Enhance Interpretation of Biodegradability Tests: A Case Study with Two Gas-to-Liquid (GtL) Products J. Dawick, G. Whale, C. Hughes, Shell Health / Risk Science Team The Oslo Paris (OSPAR) Commission, which oversees the OSPAR Convention, currently implements the harmonised mandatory control system (HMCS) for use and reduction of discharges of chemicals in the exploration and production of oil and gas offshore in the North Sea. Chemical suppliers must submit a Harmonised Offshore Chemical Notification Format (HOCNF) to the implementing OSPAR member state authority to certify use of their products offshore. Marine biodegradability screening tests (BST’s) are an intrinsic part of offshore chemical control schemes and the HOCNF registration process. However, the lack of robustness of the marine biodegradation methods has been highlighted in a series of ECETOC workshops, particularly when these are used to assess complex, volatile and poorly water-soluble substances (e.g. petroleum products). We have found that the inclusion of abiotic controls and chemical analysis for total petroleum hydrocarbons (TPH) in freshwater BST’s demonstrates that disappearance of test substances from the test system is often far greater than is suggested by the OECD guidelines and that the results are not comparable. We have presented results from two marine BST tests (OECD 306 and BODIS) conducted on two synthetic hydrocarbon Gas-to-Liquids (GtL) products. In addition to measuring ultimate biodegradation (by oxygen uptake) in these tests, additional analytical techniques have been incorporated to enhance the interpretation of results. Extraction and analysis of test samples using gas chromatography has been conducted to: i) confirm substance dosing and ii) understand the extent of abiotic losses in the test system. In addition, two-dimensional gas chromatography (GCxGC) was employed to characterise hydrocarbons present in test samples, which were compared back to the composition of the original test substances. An overview of the results and our recommendations on how marine biodegradability tests can be improved and interpreted will be provided. REFERENCES 1) ECETOC Workshop Report No.34 – Improvement of the OECD 306 Screening Test. Published September 2017. Available online via: http://www.eceto.org/publication/workshop-report-no-34-improvement-oecd-306-screening-test/ 2) Hughes, C., Whale, G., Mead, C. (2015). Investigation into the
TU288
Organising an international ring test to improve the marine biodegradation screening test
A. Graham, T. Marshall, Newcastle University / School of Engineering; G. Whale, Shell Health / Risk Science Team; J. Snape, AstraZeneca UK Ltd. / AstraZeneca Global Environment; B. Rowles, Cefas Lowestoft Laboratory; R. Davenport, Newcastle University / School of Engineering
A series of international standardised tests have been approved by the OECD to measure the relative biodegradability of substances. Among these tests, biodegradation screening tests (BSTs) form the first tier of assessment, offering relatively simple and cheap characterisations of biodegradability. Most parameters in these BSTs are highly prescribed and conservative, but the microbial inoculum is the least controlled parameter. The resulting high levels of variation have been recognised as a limitation since the introduction of these tests up to today and are especially reported for the marine BST OECD 306. BSTs were designed over two decades ago and are not, in their current form, effective as screens for persistence. In recent years, regulatory emphasis has shifted from identifying chemicals that are rapidly biodegradable to identifying chemicals that are potentially persistent in the environment. Technical guidance documents, which have been prepared under the European chemicals regulation system known as REACH, have suggested several improvements to effectively assess persistence with BSTs. Within their nature, these improvements are aimed to enhance the knowledge on the degradation of BSTs, to increase the selection of more representative test organisms and the sampling of the environment. In this study, we propose a ring test project aimed to validate and scale several improvements for the marine BST. It was possible to represent the bacterial diversity in the sampled environment better by increasing the microbial inoculum to environmentally relevant concentrations through tangential flow filtration. This standardisation led to a significant decrease in variability between replicates in comparison to the standard marine BST. By extending the test duration beyond the persistence half-life threshold, previously reported elongated biodegradation lag phases for chemicals in surface water were recognised. A ring test project was conducted from 2016-18 to validate these intra-laboratory findings from Cefic LRI ECO11 in 13 other testing facilities within Europe, North America and Japan. The test protocol, developed by academia, industry, CROs and regulatory bodies, together with information on the organisation and conduction of the ring test will be presented.

TU289
Tissue-specific accumulation of triphenyltin compounds in marine fishes in such environments as Hong Kong
R.C. Sham, K.K. Ho. The University of Hong Kong / The Swire Institute of Marine Science and School of Biological Sciences; K.M. Leung, The University of Hong Kong / The Swire Institute of Marine Science
The extensive use of organotin (OT) compounds in antifouling paints and other industrial uses (e.g. as fungicides, wood preservatives, and antibacterial textiles) have resulted in massive release of these compounds into urbanized coastal marine environments. Even though the International Maritime Organization (IMO) of the United Nations enacted a global prohibition on the usage of organotin-based antifouling agents on hulls of sea-going vessels in September 2008, Hong Kong had not adopted any regulatory legislation to restrict the production, usage and release of these compounds until early 2017. High concentrations of these compounds, especially triphenyltin (TPT), are still being detected in coastal marine environments of Hong Kong and Shenzhen, China. Organisms inhabiting these areas are particularly susceptible because they can bioaccumulate TPT through direct contact with contaminated seawater and sediment, and through dietary uptake. Nonetheless, a comprehensive tissue-specific accumulation profile of TPT compounds in marine fishes is still lacking, and such information will help reveal their toxicokinetics and identify targeted organs of accumulating these organotin contaminants. This study was, therefore, designed to investigate the distribution pattern of TPT in the bodies of four marine fish species, namely Collichthys lucidus, Cynoglossus bilineatus, Johnius belangerii, and Johnius heterolepis. For each species, 15 tissue types (n = 4) were extracted for quantification of TPT concentrations and their degradation products (i.e., di- and mono-phenyltin) using gas chromatography mass-spectrometry. We found that the accumulation tendency of TPT was highly tissue-dependent. Highest concentrations of TPT were consistently found in livers, whereas scales and swim bladders contained the least amount of TPT. Mass-balance model showed that muscles (dorsal and ventral) generally contributed to 50% of the total body burden of TPT in these fishes on a wet-weight basis. We suggested that TPT concentration of the whole organism could be predicted using its concentration in dorsal muscles (p < 0.05, r² = 0.973), which indicated that dorsal muscles can actually represent the contamination in the whole organism on dry-weight basis. Our findings from profiling the distribution pattern of TPT compounds would help identify potential TPT-induced organ-specific toxic effects in fishes, and the potential of bio-magnification of TPT in marine food webs.

TU290
POPs in the terrestrial environment of Schirmacher Hills, Antarctica: A preliminary study and implications for PCB degradation kinetics
s, katabam, IIT Hyderabad; A. Qureshi, IIT Hyderabad / Civil Engineering; P. Chakraborty, SRM University; A.K. Tiwari, NCAOR / Polar environment
We present preliminary results on the occurrence of Polychlorinated biphenyls (PCBs) and Organochlorine pesticides (OCPs) in the terrestrial environment [moss, soil, and water] of Schirmacher Hills, Dronning Maud Land, Antarctica, n-HCH compounds (4.48 ng g⁻¹ dw) and pentachlorophenol (PCP) (31.2 ng g⁻¹ dw) concentrations are higher than those observed by 5 times or more. Out of the tested 28 polychlorinated biphenyl (PCB) congeners, only 6 PCBs were detected. Σ29PCBs in moss (122 ± 115 ng g⁻¹ dw, n = 5) and water (30 ± 165 ng g⁻¹ dw, n = 2) are higher by up to 10 times compared to other studies around the continent. Heavier congeners (hexa through nona) in both moss and water samples contribute significantly to the total PCB load. This suggests that some localized sources of PCBs may still exist in the Schirmacher Hills region. It is possible that the old research stations, or tourism base, in the area may be one of those sources. While the observed congener distribution can be explained using congener distributions in known commercial PCB mixtures (Aroclors, Sovol and Clophen) in some samples, a post-deposition transformation in snow/ice (in glaciers) is required to explain the remaining observations. Box modeling exercise to reproduce congeneric distribution in our environmental samples suggests that degradation half-lives of PCB congener in snow should differ by at least 20 times between penta-/hexa-/hepta- (1750 h) and octa- (3500 h) congeners, as against the uniform value of one year (8760 h) suggested previously. Different half-lives could be possible, either if the rate of actual photoreductive-dechlorination process is different for congeners other than heptachlor. The aim of my projects which were experimentally determined previously or due to occurrence of hydroxylation reactions in snow that have been shown to be responsible for a more efficient degradation of lighter congener (PCB-7). It is also possible that the higher intensity of solar radiation in Antarctica is driving a faster degradation reaction, albeit for the lighter congeners only. Overall, PCB congener distribution in some samples can be explained by a direct contribution from sources. While for others, the post-deposition transformation step is required to reproduce the observations. More studies are required to identify and constrain the PCB sources in the Schirmacher Hills, and PCB congener degradation kinetics in snow.

TU291
Degradation of crop protection products in Brazilian soils
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Recent studies have shown that OCPs become commercially available. They are subjected to rigorous testing according to strict regulatory guidelines, including understanding the fate of these compounds in the soil environment. The global use of OCPs requires an understanding of their behaviour in a range of soils, including those from both temperate and tropical regions. Moreover, results from previous OCPs fate studies show that these compounds can be biodegraded by microorganisms. The present study was designed to determine the major physical-chemical and biological properties controlling the degradation of pesticides in Brazilian soils. A set of 4 different soils, prescribed for regulatory testing to encompass the typical range of properties, such as pH, organic matter, clay content and cation exchange capacity (CEC), including an on crop version and a pristine version of these soils, was used in my study. My first experiment focussed on the rate of degradation and mobility of the fungicide thiabendazole in four different Brazilian soils and one temperate soil. Thiabendazole exhibited slow degradation due to it adsorbing onto the soil solid surface, thus being unavailable to microorganisms in the soil pore water. Thiabendazole half-lives (DT₅₀) and distribution coefficients (Kₐₑₕ) were higher in some Brazilian soils compared to the temperate soil due differences in their physico-chemical properties. Further pesticides will be tested to determine which key physicochemical and biological properties are the driving force for a compounds fate in tropical soils.

TU292
Study of the Degradation of Bisphenol A by the basidiomycete fungus Trametes versicolor, via HPLC-DAD
C.E. Gracio, V. Bianchi, P.G. Silva, A.C. Montini, E.C. Lima, C.L. da Silva, UFABC / CCNH
Bisphenol A (BPA) is a compound widely used in polycarbonates and resins. Its use has been increasing in the last years and researches point that it may be detected in the environment in great concentrations. Moreover, this substance is classified as a pollutant of emerging concern because of its persistence in the environmental systems and its uncertain damages to both human and animal health. Some studies connect the exposition to this compound with cancer and other diseases. In this work, we evaluated the ability of the fungus Trametes versicolor (Institute of Botany of São Paulo) in degrading BPA by growing the mycelium in a enriched liquid medium and adding a Sigma brand pattern to it. After that, 2mL of sample were periodically purchased and analyzed in an Agilent 1200 Series HPLC with DAD detector. 87.78% of removal was the average efficiency of the degradation; slightly smaller than other species that our group has investigated in previous works, such as Trametes villosa. In
future works, the activity of the enzymes like Laccases and Manganese-Dependent Peroxidase should be taken into consideration and evaluated, as well as to compare the degradation with an abiotic system containing the BPA pattern – in order to evaluate its persistence in the culture medium.

TU2/203

Soil dissipation of paraffin oils: Improvement of the microbial degradation and impact on soil dissipation.

P. Adrian, A. Barret, CEHTRA SAS; G. Destrycker, CEHTRA; P. Lemaire, TOTAL Fluids

The study was conducted according to OECD 307 and the active substance applied onto soils according to Good Agricultural Practices in EU. Four soils were freshly collected from sites in Germany and handled per the International Standards Organisation Standard ISO/DIS 10381-6 Part 6 and Good Laboratory Practices.

Following incubation the soil samples are extracted and further analysed by GC/MS i.e. monitoring of one ion (m/z) for the internal standard (66 m/z) and one ion for the test substance (57 m/z). Satisfactory method performance was achieved at each degradation interval, as verified by recovery efficiency testing. The test substance, Paraffin Oil CAS (72623-86-0) as produced by TOTAL Fluids, dissipated to below 50% of the original concentration over a 17-day period after the initial treatment (DAT) in all four soils evaluated in the study. From 17 DAT through 122 DAT the concentration of Paraffin Oil CAS (72623-86-0) as produced by TOTAL Fluids, dissipated to below 80-90% dissipated in all four soils evaluated. From 122 DAT through 300 DAT there was no substantial dissipation of Paraffin Oil CAS (72623-86-0) as produced by TOTAL Fluids. Existing in any active four soils occurring. Two treated soil samples were then treated with dextrose, ammonium nitrate and monopotassium phosphate in order to improve the remaining bioactivity. The remaining residues of the applied substance was then followed for two weeks. The results showed that for one soil a complete dissipation of the remaining residues occurred although in these time of incubation a plateau was observed prior to the complete degradation. This study was therefore proposed to demonstrate for certain chemicals that the degradation is of biological origin and to integrate these results in the proposed end points.

TU2/296

Impact of biofilm growth on mercury accumulation in Daphnia magna s. issa.

M. Dancek, K. Sanchez-Thirion, LIEC; C. Crenier, LIEC; C. Mikkelsen, Norwegian University of Science and Technology / Department of Biology; S. Einum, Norwegian University of Science and Technology / Centre for Biodiversity Dynamics CBD; M. Mikkelsen, Norwegian University of Science and Technology / Department of Chemistry; V. Jaspers, Norwegian University of Science & Technology / Biology

A largely neglected issue in lab-based toxicity testing of pollutants is the potential for other components in the biotic community to influence the effects of toxins on focal species. For example, biofilms commonly grow on test organisms in such studies to infer toxicity thresholds of aquatic organisms. Nevertheless, such tests are usually conducted in highly standardized conditions and with a minimum of naturally occurring biofilm. Although this allows for standardization, it may hinder ecological relevance. Biofilm commonly grows in culture medium and serves as additional food for Daphnia. It can aslo accumulate mercury (Hg), a pollutant of high concern because of its long-range transport across the globe and its various toxic properties. As such, biofilm can play a central role in the transfer of Hg to higher trophic levels in freshwater ecosystems. By taking this into account, we can better predict effects of Hg in these ecosystems. Therefore, we conducted an experiment where single Daphnia magna clones were exposed at 20Â°C to 0.2 µg/L and 2 µg/L Hg (HgCl₂) in the presence and absence of biofilm. Our main objective was to test for a significant effect of Hg accumulation in biofilm on Hg accumulation in daphnids. Results showed no significant effect of biofilm on Hg uptake in Daphnia. However, biofilm served as an additional source of selenium (Se) to daphnids, thereby increasing Se/Hg molar ratios in the animals. Thus, biofilm played a central role in the transfer of Se through the freshwater food web and in decreasing the risk from Hg toxicity in Daphnia.

TU2/298

Multiple stressor effects on resource quality for consumers: a case study with photobiofilm exposed to phosphorus and ionic silver

M. Dancek, K. Sanchez-Thirion, LIEC; C. Crenier, LIEC; C. Mikkelsen, Norwegian University of Science and Technology / Centre for Biodiversity Dynamics CBD; M. Mikkelsen, Norwegian University of Science and Technology / Department of Biology; T.M. Ciesielski, Norwegian University of Science and Technology / Department of Biology; S. Einum, Norwegian University of Science and Technology / Centre for Biodiversity Dynamics CBD; . Mikkelsen, Norwegian University of Science and Technology / Department of Chemistry; V. Jaspers, Norwegian University of Science & Technology / Biology

Autotrophic biofilms are fundamental biological compartments of many aquatic ecosystems, representing a potentially major resource for many important consumers. To date, most studies have tried to understand the impacts of stressors on microbial communities or on functional processes taking place into the biofilm. For example, studies investigating the indirect effects of stressors on upper trophic levels through alterations of the quality of biofilms. We investigated, through a laboratory study, the single and combined effects of phosphorus (P) availability and silver contamination on the elemental (Ca, P, Na, K) and biochemical (fatty acid profiles) compositions of a diatom-dominated biofilm. We hypothesized that 1) P would enhance the elemental quality while 2) P and silver, through the replacement of diatoms by more tolerant primary producer species, would reduce the

When ecotoxicology meets trophic ecology (P)

TU2/205

Will detoxification processes of marine mammals still be effective in the future?

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In marine mammals, food is the main route of entry for contaminants. Their concentrations can largely vary among prey species, for that reason differences in bioaccumulation will arise from differences in predator diet. Among all the contaminant that marine organisms faces, metallic trace elements (MTE) are natural substances that have been present on the earth since its formation. MTE can be divided in essential and non-essential in function of their biological role in the organisms. Low concentrations of essential elements can lead to deficiency effects. On the contrary, excess of non-essential elements (i.e. cadmium (Cd), mercury (Hg) and lead (Pb)) can induce toxic effects. However, their long-term presence in to the environment has allowed to marine mammals and other marine organisms to developed mechanisms to mitigate the potential toxic effects of these non-essential elements. The best known detoxification process is the demethylation of Methyl-Hg by Selenium (Se) forming granules of tiemmanite (Hg₅Se₆) in their liver.

Today, anthropogenic activities induced a continuous increase of Hg concentrations in the environment, altered the geographical distribution of Se and its various communities. Such changes could affect the ability of marine mammals to control the negative impacts of their exposure to non-essential elements. Here, we investigated the temporal trends of Hg and Cd in liver and kidneys (main storage tissues) of 183 individuals of the smallest cetacean species in the North Atlantic: the harbour porpoise (Phocoena phocoena). Both elements showed a significant increase (p < 0.05) of concentrations between 1999 and 2013. Notably, we highlighted a strong increase of the number of individuals exhibiting extreme values among the range of measured concentrations. In parallel, we analysed essential trace elements in 78 forage species (i.e. jellyfish, crustaceans, cephalopods and cartilaginous and bony fish) to assess their quality for predators. Results showed broad differences of their essential element composition. In particular, selenium concentrations can be low because of its long-range transport different Se exposure among marine mammal species depending on their diet, which means that some of them could be less protected against Hg toxicity. Thus, changes in prey quality could have cumulative effects in cetaceans (increase of toxic elements and deficiency in essential ones) impacting the efficiency of detoxification processes in the future.

TU2/295

Multiple stressor effects on resource quality for consumers: a case study with photobiofilm exposed to phosphorus and ionic silver

M. Dancek, K. Sanchez-Thirion, LIEC; C. Crenier, LIEC; C. Mikkelsen, Norwegian University of Science and Technology / Department of Biology; T.M. Ciesielski, Norwegian University of Science and Technology / Department of Biology; S. Einum, Norwegian University of Science and Technology / Centre for Biodiversity Dynamics CBD; . Mikkelsen, Norwegian University of Science and Technology / Department of Chemistry; V. Jaspers, Norwegian University of Science & Technology / Biology

A largely neglected issue in lab-based toxicity testing of pollutants is the potential for other components in the biotic community to influence the effects of toxins on focal species. For example, biofilms commonly grow on test organisms in such studies to infer toxicity thresholds of aquatic organisms. Nevertheless, such tests are usually conducted in highly standardized conditions and with a minimum of naturally occurring biofilm. Although this allows for standardization, it may hinder ecological relevance. Biofilm commonly grows in culture medium and serves as additional food for Daphnia. It can aslo accumulate mercury (Hg), a pollutant of high concern because of its long-range transport across the globe and its various toxic properties. As such, biofilm can play a central role in the transfer of Hg to higher trophic levels in freshwater ecosystems. By taking this into account, we can better predict effects of Hg in these ecosystems. Therefore, we conducted an experiment where single Daphnia magna clones were exposed at 20Â°C to 0.2 µg/L and 2 µg/L Hg (HgCl₂) in the presence and absence of biofilm. Our main objective was to test for a significant effect of Hg accumulation in biofilm on Hg accumulation in daphnids. Results showed no significant effect of biofilm on Hg uptake in Daphnia. However, biofilm served as an additional source of selenium (Se) to daphnids, thereby increasing Se/Hg molar ratios in the animals. Thus, biofilm played a central role in the transfer of Se through the freshwater food web and in decreasing the risk from Hg toxicity in Daphnia.
biochemical quality of biofilms for their consumers. The quality of biofilms for consumers was assessed for a common crustacean species, *Gammarus fossarum*, by measuring organisms survival and growth rates. Results mainly showed that species replacement induced by both stressors affected biofilm fatty acid compositions, and that P immobilization permitted to achieve low C/P biofilms, whatever the level of soil contamination. Gammarids growth and survival were not significantly influenced by the ingestion of silver-contaminated resource. On the contrary, we found a significant positive relationship between the biofilm P-content and the growth of gammarids, while biofilm fatty acid contents were unrelated to this parameter. This study underlines the large indirect consequences stressors could play on basal resources quality for consumers, and, in turn, on the whole food web.

TU309

Soil pollution induced changes in leaf litter chemical composition and in detritivore physiology and activity.

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In terrestrial ecosystems contaminated with high metal contents (brownfield), recent studies showed that, surprisingly, leaf litter decomposition process could be maintained despite deep changes in bacteria to fungal abundance ratio and invertebrate detritivore community structures. To disentangle the potential mechanisms leading to this pattern, we e

TU300

Decomposition rates and feeding activity of soil fauna in relation with stages of plant colonization in mine soils of a Mediterranean area


Organic matter decomposition (tea bag index) and feeding activity of mesofauna (Bait Lamina) were studied in an abandoned mine tailings area. Six environments were studied: A) Within the mine tailings: 1. Bare soils (S); 2. Small groups of Pinus halepensis trees (2-5) ≤2.5 m high, growing scattered (P); 3. Isolated P. halepensis tree (5) ≤2.5 m high with shrubs under the canopy (DP+MS); 4. Dense patches with several P. halepensis trees (5) >4-5 m high and shrubs and herbs under the canopy (DP+MS). B) Outside the mine tailings: 5. Polluted forest with P. halepensis trees >5 m high and shrubs and herbs under the canopy (CF); 6. Control forest not contaminated with P. halepensis trees >5 m high and shrubs and herbs under the canopy (CF). Roibos and green tea-bags were buried in each environment, for a period of 110 days. Tea-bags exposed to each environment, to calculate mass remaining, decomposition index and organic carbon and nitrogen of the remaining material. In each plot, two groups of 5 baited sticks were vertically inserted. The number of holes partially and fully empty after 20 days was recorded to calculate the % of holes fed upon. After >50 days, the percentages of mass remaining in the tea bags were: DP+MS, 24.9%; CF and P. green tea =50-55%, roibos tea =90%; PF, CF and P. green tea =80-85%, roibos tea =90%. These percentages were maintained until the day 110. The lower decomposition in CF and PF can be related with more abundance of resources in forest soils outside of the tailings which could induce to microorganisms to use other sources of nutrients different from tea material. However, in more stressed environments, such as mine soils with a very resting resource, decomposition is easily controllable, mainly the green tea, could stimulate microbial activity by a priming effect. Besides, within the tailings, the decomposition in S (the most unfavorable environment "a priori") could be favored by the high soil temperature (average ≈28°C), as a consequence of the lack of vegetation, while in other environments the shading by plants maintained the temperature between ≈23 and ≈25°C. Feeding activity was ≈5% of holes fed upon. CF =32%, P. 31%, PF =31%, CF =8%, DP+MS =7%. The high % of holes fed in bare soils (S) could be related to the scarcity of resources in this environment which stimulated the consumption of the bait.

TU301

Effects of mineral supplements on leaf exposure in free-ranging herbivores

J. Pareja Carrera, IREC-UCLM / IREC-UCLM; M. Martinez-Haro, IREC-Instituto de Investigación en Recursos Cinegéticos / Department of Life Sciences; J. Rodr

University of Calgary / Ecosystem and Public Health; M. Durkalec, National Veterinary Research Institute / Department of pharmacology and toxicology; R. Mateo, IREC-CSIC - UCLM / Grupo de Toxicología de Fauna Silvestre

Lead (Pb) mining has contributed to the extensive release of Pb into natural environments for centuries. In former mining districts, now on the Iberian Peninsula mainly livestock and hunting estates, Pb persisting in the soils and vegetation of affected areas may contribute an environmental and health risk. Since Pb is a toxic metal for both animals and people, there is a need to explore how to prevent or reduce exposure. Therefore, we studied the effect of commercial mineral supplements on Pb bioaccessibility through laboratory and field approaches. One aim was to prevent or reduce absorption in herbivore inhabiting mining areas and thus reduce the possible exposure route to people. In our previous work, we identified one mineral supplement rich in calcium (Ca) and phosphorus (P) that specifically altered Pb solubility and absorption in a digestive tract simulation model. Here, we go one step further to evaluate the effects of that commercial mineral supplement on Pb absorption and on immune status in goats from an old mining area. Two groups of goats from two plots with similar soil Pb concentrations were selected. One group was supplemented with the commercial mineral salt for 20 days, whereas the other one served as control. Then, the Pb exposure was evaluated in blood, milk and feces, and the phytosiderophilin test was used to evaluate T-cell mediated immune competence. Results showed that all goats responded to the PHA, but no significant difference was detected between groups. Blood and milk samples were collected the same days that the PHA skin test was conducted. Blood Pb levels in supplemented goats were 2-fold higher than in supplemented animals (0.012 vs. 0.006 µg/g wet weight). Based on our study, supplements enriched with Ca and P appear to decrease the Pb burden in the animals, likely through competition in intestinal absorption with the nutrient, Ca, preferentially absorbed over the toxic metal, Pb, and possibly through reduced geophagia by the animal. The experiment was repeated with a different animal stock, as both plants and wildlife in areas contaminated with Pb. An assumed additional advantage is reduced the risk of Pb exposure through milk consumption by the local human population.

TU302

Analysis of anticoagulant rodenticides, neonicotinoids and fipronil in liver of predatory birds


Pesticides in predatory birds have been drawing much attention worldwide in regard to species declining and protection. Pesticides are used for pest management of animal species such as commensal rodents and sap-sucking insects. However, pesticides can lead to secondary poisoning, when rodents take up pesticides and these residues from primarily exposed target or non-target species. The analysis focused on anticoagulant rodenticides, neonicotinoids and fipronil which were regularly applied in the years 2011 to 2013. We obtained liver samples of 89 avian predators from this period, which were collected from veterinary institutions or private persons from 26 administrative districts in Germany. Avians were found dead or dead shortly after admission to the veterinarian. Defrosted liver samples were spiked with surrogates and homogenized in a mixture of methanol and water (2:1/v/v) and cleaned up by solid supported liquid extraction with a diatomaceous earth column (Geduhn et al., 2014, DOI 10.1016/j.scitotenv.2014.07.099679). Quantification of the analytes was performed by LC-ESI-MS/MS with a calibration from 0.1 to 100 ng/ml (r² > 0.99) and a signal to noise ratios > 6.1 for the lowest concentration level. The neonicotinoids imidaclopid with the metabolites 5-OH-IMD and IMD-olefine, thiamethoxam and clothianidin with TZMU and TZNG were not found in the predators although expected especially in case of insect-consuming species such as little owl (*Athene noctua*). Similarly, we detected no residues of the phenytoilamid fipronil, which has a higher bioaccumulation potential and the metabolites, F-carboxamide, F-sulfide and F-carboxamide. One to four substances of the rodenticides chlorophacinone, difencafen, bromadiolone, brodifacoum, flocoumafen and diphenthiamine were found in 30% of the liver samples, originated from 14 different districts. Brodifacoum was detected in more than 70% of these samples. No sample contained coumatetralyl and warfarin. Residues occurred more often in avian species specialized on rodents than in generalists; e.g. 44% of the 26 liver samples from common buzzards (*Buteo buteo*) contained residues. The portion was with 80% even higher for red kite (*Milvus milvus*) but only 5 samples of this species were examined. A residues distribution pattern will be presented but more samples are necessary for final statements.

TU303

Trophic Magnification of POPs including PFCs Within A Terrestrial Food-Web Of An Avian Top Predator, the Cooper's Hawk (Accipiter Cooperii)

303

SETAC Europe 28th Annual Meeting Abstract Book
Bioaccumulation of POPs within terrestrial systems is far less developed compared to aquatic systems. Presently, regulatory agencies in Canada, the USA, and the EU use only bioaccumulation information for fish to assess the bioaccumulation potential of chemicals. However, recent studies have shown that some chemicals that are not biologically accumulated in aquatic food-webs do biomagnify in terrestrial food-webs. To better understand the bioaccumulation behaviour of chemicals in terrestrial food-webs, we aim to produce a food-web model to assess the bioaccumulation of POPs in an apex avian predator, the Cooper’s hawk. Over 100 samples were collected from various trophic levels of the food-web including hawks, eggs, songbirds, invertebrates, and berries. All samples were analysed for a number of contaminants listed as priorities for monitoring by the Chemical Management Plan of the Canadian federal government. Stable isotope analysis of δ15N and δ13C signatures of hawks, songbirds, invertebrates, and berries was used to estimate the trophic position of each organism. Legacy POP concentrations were expressed in terms of lipid equivalent concentrations to account for variability in the fractions of lipid and non-lipid organic matter measured in each sample. PFC concentrations were expressed in terms of protein equivalent concentrations to account for the fraction of protein within each sample, which was estimated as the product of the percent of nitrogen measured in each sample and a nitrogen:protein conversion factor. Censored regression by maximum likelihood estimation was used to assess the relationship between the natural logarithm of each lipid or protein equivalent concentration and visual trophic position. Trophic magnification factors (TMFs) were determined as the antilog of the regression slope. TMFs of legacy POPs ranged from 0.61 to 38.40, indicating that most legacy POPs are biomagnifying in this terrestrial food-web. TMFs of PFCs ranged from 11.8 to 544.6, indicating that PFCs are also biomagnifying in this terrestrial system and potentially at higher magnitudes than legacy POPs. Overall, terrestrial TMF values for PFCs were determined to be similar for several aquatic systems; whereas, terrestrial TMF values for the PFCs were considerably higher than TMF values found in aquatic systems.

**TU306** Comparative trophodynamics of polychlorinated biphenyls and chlorinated paraffins in an urban river basin. 
K. Fremlin, SFU / Department of Biological Sciences; J.E. Elliott, Environment Canada / Science Technology Branch; F. Gobas, Simon Fraser University / Resource & Environmental Management

Protocols to assess bioaccumulation of POPs within terrestrial systems are far less developed compared to aquatic systems. Presently, regulatory agencies in Canada, the USA, and the EU use only bioaccumulation information for fish to assess the bioaccumulation potential of chemicals. However, recent studies have shown that some chemicals that are not biologically accumulated in aquatic food-webs do biomagnify in terrestrial food-webs. To better understand the bioaccumulation behaviour of chemicals in terrestrial food-webs, we aim to produce a food-web model to assess the bioaccumulation of POPs in an apex avian predator, the Cooper’s hawk. Over 100 samples were collected from various trophic levels of the food-web including hawks, eggs, songbirds, invertebrates, and berries. All samples were analysed for a number of contaminants listed as priorities for monitoring by the Chemical Management Plan of the Canadian federal government. Stable isotope analysis of δ15N and δ13C signatures of hawks, songbirds, invertebrates, and berries was used to estimate the trophic position of each organism. Legacy POP concentrations were expressed in terms of lipid equivalent concentrations to account for variability in the fractions of lipid and non-lipid organic matter measured in each sample. PFC concentrations were expressed in terms of protein equivalent concentrations to account for the fraction of protein within each sample, which was estimated as the product of the percent of nitrogen measured in each sample and a nitrogen:protein conversion factor. Censored regression by maximum likelihood estimation was used to assess the relationship between the natural logarithm of each lipid or protein equivalent concentration and visual trophic position. Trophic magnification factors (TMFs) were determined as the antilog of the regression slope. TMFs of legacy POPs ranged from 0.61 to 38.40, indicating that most legacy POPs are biomagnifying in this terrestrial food-web. TMFs of PFCs ranged from 11.8 to 544.6, indicating that PFCs are also biomagnifying in this terrestrial system and potentially at higher magnitudes than legacy POPs. Overall, terrestrial TMF values for PFCs were determined to be similar for several aquatic systems; whereas, terrestrial TMF values for the PFCs were considerably higher than TMF values found in aquatic systems.

**TU307** Copper and mercury effective body residues in freshwater macroinvertebrates as related to benthic community metrics from a mining river basin.

Copper and mercury body residues in 10 macroinvertebrate taxa were used to model the alterations in benthic community metrics due to metal bioaccumulation in mining areas of the Nalón River basin (Spain). The studied taxa are potentially useful as water quality biomonitor and cover different functional feeding stages. This is part of a larger study in North Spain that aims to develop bota quality...
in the digestive gland revealed diverse lesions ranging from the l/n loss of cilia and covering membranes, to the increase in the light of the digestive gland tubules, as well as the presence of various/inflammatory processes. Other organs such as the gills, presented/inflammation and injuries that compromise the body’s physiological processes/in such as feeding and breathing. These damages were evident after the first 96/h of exposure to the contaminated food. However, lesions derived from copper and cadmium exposure, a non-essential metal, in more than 50% of organisms/in could be observed on day 10 and those associated with more than 50% ‘n animals in coo exposure were deferred to day 15. The presence of Chlorella/in sp. in the digestive tract made possible to associate the injuries within trophic metal poisoning, and the prevalence of lesions with metal and exposure time.

TU310 Can microplastics save us? Effects of microplastic particles and particle-bound trace contaminants in an artificial aquatic food web

I. Hanslik, COS University of Heidelberg / Aquatic Ecology and Toxicology; A. Batel, University of Heidelberg / Aquatic Ecology and Toxicology; T. Brauneck, University of Heidelberg / Centre for Organismal Studies.

Since 2015, studies dealing with toxic effects of microplastics in freshwater ecosystems come into focus. Still, little is known about vertebrate models as final consumers in food web experiments. The present study investigates the transfer of pristine microplastic particles and a model polycyclic aromatic hydrocarbon, along an artificial food chain with Artemisia spec. nauplii and zebrafish (Danio rerio). Therefore, cryo-technically grinded microplastic particles, made of polystyrene (P)

TU311 Toxicokinetics links predator-prey dynamics to assess zero-valent iron nanoparticles bioaccumulation in a Caenorhabditis elegans-Escherichia coli ecosystem

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BACKGROUND: Zero-valent iron (Fe0) nanoparticles (NPs) are one of the most paramount NPs applied in environmental remediation that the potential impacts on the ecological dynamics and soil ecosystem health are of great concern. OBJECTIVE: The primary objective of this study was to simulate dynamic models linking bioinert and consumer-resource dynamics in the Caenorhabditis elegans (C. elegans)-Escherichia coli (E. coli) OP50ecosystem. METHODS: The bioinert parameters, uptake and depharosorption rate constants of bacteria and worms were obtained from toxicokinetic experiments and related published literature. Biomass dynamics of bacteria and worms were estimated by employing the Lotka-Volterra model. Dynamics of Fe0/NPs accumulations, bioconcentration factors (BCFs), biomagnification factors (BMFs) were simulated based on the consumer-resource dynamics. A sensitivity analysis was also performed to characterize the influence of consumer-resource-related physiological parameters. RESULTS: Results showed that biomass of worms increased steadily from 22.25–51.61 g L−1, whereas the biomass of bacteria decreased rapidly from 17.17–2.29 g L−1 and attained a steady-state after 2 h of the simulation in the scenario of 100 mg L−1 Fe0/NPs exposure. We also observed that internal concentrations of Fe0/NPs were estimated to be 67 and 1768.85 µg L−1 in worms and bacteria, respectively. In addition, the BCF of bacteria was found to be 17.69, close to the experimental results. Moreover, the BMFs of worms were maintained to be consistently smaller than 1 during 24 h exposure. Results also indicated that internal concentrations of Fe0/NPs in worms were mainly influenced by biomass conversion rate for bacteria ingested by worms, whereas parameter of death of worms had the slightest effect on worm internal concentrations. CONCLUSIONS: Model application to toxicokinetic results confirms the hypothesis that the consumer-resource dynamics are effectively associated with Fe0/NPs accumulations in bacteria and worms that the bioaccumulation kinetics and consumer-resource dynamics are likely to be dominated by the same physiological parameters.

Use of Effect Based Methods in the context of the national and european legislative framework for the protection of aquatic ecosystems (P)

TU312 INTEREST OF IN VITRO BIOASSAYS (YES/YAS) FOR THE SCREENING OF ENDOCRINE DISRUPTION IN SURFACE WATERS OF WOLLONIA (BELGIUM)

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This study is part of the BIODIEN project. This project aimed at conducting, for the first time, a screening campaign of endocrine disruptors (ED) in waters of Wollonia (groundwater, surface water and wastewater). Almost 200 substances were screened, including hormone estrogen, alklyphenols, phthalates, chlorophenols,
perfluoranes, PBDEs, PCBs, HAPs and pesticides. In parallel with analytical methods, YES and YAS bioassays were conducted in order to quantify estrogenic and androgenic activities in surface waters. Antagonist activities were also evaluated. Over 71 river samples (concerning 24 river sampling points from the regional monitoring network), estrogenic activity was detected and quantified in 53 samples and could reach levels up to 11.7 ng E2eq/l (mean: 2.1±1.6 ng E2eq/l). Androgenic activity was detected. At the other hand, estrogenic and antagonist activities were detected in 42 % and 55 % of the samples, respectively. When the estrogenic activity is compared to the EU-Watch List EQS for E2 (0.4 ng/l), 60 % of the samples exceed this value. The estrogenic activity was compared to the chemical results. A good correlation was found with the estrone concentration but also with other ED (e.g. bisphenol A, perfluoranes). This study is, in a way, the first attempt in Wallonia to follow the recommendations for the use of effect-based methods (EBM) for monitoring of estrogenic in surface waters emanating from the Science to Policy Interface (SPI) Estrogen monitoring project. These recommendations were presented at the last EU-WG chemicals held in October 2017 and this would possibly lead to the introduction of EBM in regulatory monitoring under the Water Framework Directive (WFD), especially for estrogens.

TU313 Ecotoxicological tools to assess the impact pollution of tributaries to the Alqueva Reservoir (Southern Portugal)

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As part of an ongoing review of the Water Framework Directive (WFD), the European Commission (EC) is considering “effects based tools” (EBTs) for use as an alternative to monitoring. The first step is to perform a literature review on the use of effect-based approaches for the use of effect-based methods (EBM) for monitoring of endocrine disruptors in surface waters. There are several ways in which EBTs may be integrated into operational programmes, and propose a list of assays for use in monitoring surface water concentrations. These assays were then broadly screened based on commercial availability, general validation maturity, previous application to environmental samples, and suitability for use with passive sampler extracts to derive a short list of 22 assays for more detailed consideration. The short-listed assays included novel whole organism bioassays (or surrogates), and in vitro or bacterial assays for endpoints based on endocrine disruption (oestrogen, androgen and thyroid), genotoxicity, oxidative stress, and metabolism of polyaromatic hydrocarbons (PAHs). Commonly used whole-organism assays (e.g. aequorin, algal growth inhibition and Daphnia magna) were considered as part of the final recommendations. The shortlisted EBTs were then subject to a detailed review, based on the published scientific literature, to identify relevant information with respect to their performance, interpretation, and application. The EBTs were compared using the information identified in the literature review and an initial suite of thirteen bioassays were recommended for the monitoring of surface waters associated with refinery effluents using passive sampler extracts. This recommended suite of EBTs should be considered a starting point for use in the monitoring of waters receiving refinery effluents, to be further developed based on experience in using the assays for this purpose, and incorporating new relevant bioassays once they have achieved a sufficient level of validation maturity. This review will be published as a Concawe report in 2018.

TU315 Ecotoxicological monitoring techniques for the protection of aquatic ecosystems and the implementation of the Water Framework Directive (WFD)

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The Water Framework Directive (WFD, 2000/60/EC) regulates the European water bodies. The Directive aims at achieving the Good Environmental Status (GES) of the aquatic ecosystems and the implementation of the Water Framework Directive (WFD) policy and addresses the EU member states to achieve good qualitative and quantitative status of all water bodies. Despite the efforts to reduce the release of chemicals into the aquatic environments, pollution is still widespread across Europe, and new emerging substances should be assessed and managed. The general goal of this project (realised in the framework of the ‘Torno subalpino Lazio Operational Programme Europe Social Funding 2014-2020) is to select and define innovative methods to assess the toxicity due to the exposure to different pollutants, especially the emerging substances and respective mixtures, with a focus on aquatic ecosystems and human health. This goal has been achieved, in a first step, by making a literature review on the priority and emerging substances widespread in the aquatic environment, to investigate their effects on the development of zebrafish (Danio rerio) embryos. Then, a few toxic substances that are relevant for our goals have been selected and analysed through the fish embryo acute toxicity test (FET) and other assays; particular attention has been given to the sub-lethal effects. Afterwards, environmental samples from different aquatic systems in Italy will be analysed to detect the chemicals present in these sites as well as their toxic effects. In order to reach a better comprehension of the effects of some emerging substances on the development of fish, a battery of fish tests (bacteria assays for endpoints based on endocrine disruption (oestrogen, androgen and thyroid), genotoxicity, oxidative stress, and metabolism of polyaromatic hydrocarbons (PAHs). Commonly used whole-organism assays (e.g. aequorin, algal growth inhibition and Daphnia magna) were considered as part of the final recommendations. The shortlisted EBTs were then subject to a detailed review, based on the published scientific literature, to identify relevant information with respect to their performance, interpretation, and application. The EBTs were compared using the information identified in the literature review and an initial suite of thirteen bioassays were recommended for the monitoring of surface waters associated with refinery effluents using passive sampler extracts. This recommended suite of EBTs should be considered a starting point for use in the monitoring of waters receiving refinery effluents, to be further developed based on experience in using the assays for this purpose, and incorporating new relevant bioassays once they have achieved a sufficient level of validation maturity. This review will be published as a Concawe report in 2018.

TU316 Chemical and Ecotoxicological Monitoring of a marine coastal area in the Central Italy

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A monitoring campaign in Central Italy with the aim to characterize the chemical quality status of the coastal marine area in order to detect the possible impact of the emissions of a Coal fired power station and other sources of pollution in proximity of the city of Civitavecchia. The sampling has been carried out in two different seasons of the year along the marine coastal area and in a transitional surface waterbody (Saline di Tarquinia). The analysis has been performed on the water column and at the first 20 cm of the sediments. The chemical substances analyzed included several priority substances of the WFD (water framework directive) and other chemical substances: Metals, Dioxins, PCB, PAH, Naphtalene. The Ecotoxicological assays have been performed with the use of algae (Phaeodactylum tricornutum) and crustaceans (Artemia franciscana and Tigriopus fulvus). The results have showed a diffuse light exceedance of the sediment environmental quality standards of the Italian legislation for some metals (e.g. arsenic, lead, chromium, mercury) and naphtalene; the data of the water column are in general below the environmental quality standards, but Uranium has been detected in surface water samples at concentrations above the available PNEC
(predicted no effect concentration). Ecotoxicological effects have been detected with the algae and could be related to the substances detected (e.g. heavy metals) or other substances released in the area (transitional waterbody). In general the results show a situation in which the quality of the sediments is not in a good status, although the level of concentrations should not cause a high risk for the aquatic ecosystems; the chemical contaminants can derive from different sources of pollution (industrial, urban, agricultural, atmospheric deposition) and might be reinforced due to the poor quality water required by the water framework directive. The presence of Uranium in the marine coastal area should be further investigated to understand the possible role of the Coal fired power station.

TU317 USE OF DIAGNOSTIC STRAINS OF THE SALMONELLA/MICROSOME ASSAY FOR THE IDENTIFICATION OF MUTAGENIC PROFILES IN WATER SAMPLES AND SUSPENDED PARTICULATE MATTER D.A. Morales, State University of Campinas / Faculty of Technology; J. Rossetto Martins Zwarg, School of Technology, UNICAMP; R. Massei, Helmholtz Centre for Environmental Research UFZ; T.C. Schmidt, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis; G. Umbuzeiro, School of Technology, UNICAMP / LEAG

The Salmonella/microsome mutagenicity assay uses the genetically modified Salmonella enterica serovar Typhimurium bacterium, and the exploration of these different genetic characteristics allows the detection of various classes of mutagenic compounds. The objective of this study was to use different strains for the identification of mutagenic profiles and mixing at the level of compounds responsible for the detected mutagenicity. The research was performed with organic extracts of surface water and suspended particulate matter (SPM) collected in 13 different sites along the Danube River during the Joint Danube Survey 3 (JDS3). The extracts were evaluated with the Salmonella/microsome microsuspension activation assay and the comet assay (TAX, YG1041, TA1538 and YG5185 with specific genetic characteristics for the detection of different compounds. A total of 69% surface water and 92% SPM samples were positive for at least one strain-condition. The applied methodology compared the profiles obtained at different sites along the Danube River. In this way, it was possible to classify different types of contaminated water samples that are causing mutagenicity. We conclude that aromatic amines and nitro compounds for surface water and also polyyclic aromatic hydrocarbons for suspended particulate matter appear to be responsible for mutagenicity at some of the sites, because the obtained profile were similar representative compounds of these classes. Other sites indicate the presence of other types of compounds or mixtures that need to be investigated. Non-target analysis is currently carried out to test if it is possible to correlate the observed profiles with individual compounds or groups of chemicals.

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TU318 NTA meets EDA: A practical example J. Kuijke, V. Hinnenkamp, P. Balsaa, A. Simon, IWW Rheinisch-Westfälisches Institut für Wasserwirtschaft GmbH; T.C. Schmidt, University of Duisburg-Essen

Organic micropollutants play an important role in the assessment of water bodies that are used for drinking water production. On one hand, micropollutants pass through the wastewater system in sewage treatment plants and subsequently in surface water. On the other hand, there are direct discharges from industry and in addition, there are diffuse sources from agriculture, or from households, buildings and settlements. As major pollution events (i.e. PPC in the river Ruhr) show, water suppliers must always expect to find new critical substances that could pose a potential health risk in drinking water, especially when using surface water. For this reason, it is necessary to initiate a proactive screening of contaminants and their potential effects. Continuous monitoring by high-resolution mass spectrometry also makes it possible to analyze pollution retrospectively, gain knowledge about temporal dynamics and discharge patterns and thus identify the source of the contamination more frequently. In this context, a fast, robust and routine method for the determination of organic micropolllutants is needed. This project is an approach to analyze organic micropolllutants in water samples with a combination of non-target-analysis (NTA) and effect-directed-analysis (EDA). Samples were taken regularly over a period of one year in order to obtain an annual progression of the water pollution. A LC MS QTOF system was used to carry out the NTA. Different endpoints were analyzed for the EDA: cytotoxicity (MTT assay), endocrine activities (ER-CALUX and AR-CALUX) as well as genotoxicity (p53-CALUX, umuC assay and Ames assay). Due to the investigation of raw water samples, no significant biological effect of the individual samples was to be expected. The focus of the project was therefore to detect seasonal experiments of the micropolllutant load. The coupling of NTA with a test strategy for toxicological effects forms an innovative approach with potential for preventive product quality assurance of the water supplier.

TU319 Imposex levels in gastropods from the Northern Adriatic Sea (Italy): a proposal of classification according to the Water Framework Directive P. Cacciatore, ISPRA-Institute for Environmental Protection and Research / Mitigation and Prevention of Impacts; R. Boscollo Brusà, C. Antonini, M. Femmerlewicz, ISPRA - Institute for Environmental Protection and Research; M. Marin, University of Paduaa; A. Bonfiglio, M. Gabelini, ISPRA - Institute for Environmental Protection and Research

Butyltins (BTs) - i.e. mono- (MBT), di- (DBT) and tributyltin (TBT) - are synthetic compounds worldwide used in industrial and agricultural applications giving rise to contamination of aquatic environments. Organotins, which include BTs, are banned on antimouling paints, the main route to aquatic pollution, by Reg. 782/2003/EC. Presently, due to their persistence, toxicity and bioaccumulative properties, TBT compounds are included among the priority hazardous substances according to the European Water Framework Directive (WFD) and its daughter Directive 2008/105/EC. Imposex, the superimposition of male sexual characteristics on females of gonochoristic gastropods, is the most studied effect of TBT and it is generally recognized as a water quality and aquatic ecotoxicity biomarker. For this reason, under the implementation of the WFD, imposex measurements have been indicated as a useful tool to link chemical and ecotoxicological status assessment. In this context, the aim of this study was to propose a classification of imposex according to WFD using two species of gastropods collected in the Northern Adriatic Sea and in the Venice Lagoon: Nassarius nitidus (Jeffreys, 1867) and Hexaplex trunculus (_predicted no effect concentration). Ecotoxicological effects have been detected with the algae and invertebrates. The challenge at this time was to define Ecological Quality Ratio (EQR) class boundaries within WFD, the relationship between the ecological impact caused by BT pollution and the reproductive capacity of the gastropod populations was assessed. This preliminary attempt showed that most of the sites were in Bad ecological status before the ban and have reached mostly a Moderate status at present, with very few sites in Good or High status. A comparison between the two species was also performed showing advantages coming from the combined use of both species to cope with the ecotoxicological status assessment in a wider study area.

TU320 Lessons Learned from Sibro Dam and River Restoration in Sweden E. Hallqvist, C. Becker, P. Bönlöcke Adansson, P. Gliveson, A. Sahlen, Ramboll Aquatic ecosystems in the European Union are under pressure from growing demand for insufficient quantities of good quality water for human use. The Water Framework Directive (WFD) enforces the improvement of the ecological status of rivers and lakes, and it is directly linked to the ecological quality standards (EQS) that have been established as legal tools with which to set requirements for member states. In Sweden, all major surface waters are classified according to the current status of the water designated by authorities in the respective water district. The ecological status of surface water comprises three different types of quality factors according to the WFD: biological, physical and chemical and hydro-morphological. The latter defines connectivity and biodiversity in the ecosystem, since many aquatic organisms are independent on the ability to migrate during their life cycle. Water power represents a large fraction (almost 50%) of electricity production within the country, and a large proportion of Swedish rivers are affected hydro-morphologically. At present, there are 11,000 active and abandoned dams in Swedish rivers, and 1,801 are hydroelectric power plants. All of these dams impact the ecological connectivity of rivers and have a negative impact on biodiversity. In Sweden, a common national strategy is to increase the use of hydropower/plants as an alternative to reliance on fossil fuels. In the same time water power is the greatest individual cause of physical impacts in lakes and streams. The challenge at this stage is defining what Sweden’s national energy strategy is to identify technologies and management practices that promote hydroelectric power with minimal long-term adverse/ecological impacts. To illustrate the challenges, this paper summarizes work conducted in Sweden over the past 2 years to manage the future of the Sibro Dam located in southern-central Sweden. The project was initiated after previous dam repair work involving the diversion of large amounts of the Sibro River aimed to protect and improve consequences for nationally protected indigenous mussels and other aquatic life. The responsible municipality is obligated to improve ecological connectivity at Sibro Dam and regulation of Lake Bäven. The planning/implementation of a proposal of environmental impact assessment (EIA), detailed engineering design for fish passage, engagement with local communities and communication between the municipality of Nyköping and Sweden’s federal court in Keywords/Fish passage; Sweden; ecological connectivity; environmental impact assessment

TU321 Behavioural Ecotoxicology: Unravelling behavioural responses to chemical contaminants in the environment (P)

Behavioural Ecotoxicology: Unravelling behavioural responses to chemical contaminants in the environment (P)
Marine and estuarine fish accumulate methylmercury (MeHg) to elevated concentrations, often higher than in freshwater systems. Because MeHg is a neurotoxin, it is plausible that high tissue concentrations could affect behavior in marine fish which in turn could affect their populations in contaminated waters. However, we measured behavioral effects of MeHg to a marine forage fish at the larval stage, the Sheephead minnow Cyprinodon variegatus. Because the bioavailability of MeHg from different food types may lead to different MeHg internal distributions and toxic effects, we compared artificial and natural diets with varying MeHg concentrations. Artificial (commercial fish flake containing methylmercury) or natural diets (zooplankton containing MeHg, obtained from MeHg-contaminated phytoplankton) were prepared; MeHg concentrations ranged from zero (controls) to as high as 7.8 ppm. The larvae were fed control and MeHg-contaminated diets from an age of 7 days until 5 weeks when they reached juvenile stage. Growth rates, respiration rates, and swimming activity were tested. Results indicate that MeHg-rich diets—either artificial or natural foods—have no significant impact on fish growth rates under any treatment. However swimming activity (swimming speed, acceleration, active time and swimming distance) was impaired after 3 weeks exposure to natural diets containing 7.8 ppm but not 2.5 ppm; artificial diets containing MeHg up to 4.8 ppm had no discernible effect on swimming. In addition, MeHg as low as 4.8 ppm had a small but significant impact on the respiration rates of these fish. The data suggest that mortality and growth are unlikely to be affected by these MeHg concentrations, but swimming activity may be reduced at elevated MeHg concentrations, and this could influence the success of populations in the wild through impairment of predation or avoidance of predators.

TU324

Impacts of environmentally realistic antidepressant exposure on reproductive behaviour and sperm traits in fish

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Pharmaceutical contaminants are increasingly being detected in ecosystems worldwide. Indeed, more than 1 in 10 currently manufactured pharmaceuticals have been found in the environment. One pharmaceutical pollutant of environmental concern is the antidepressant fluoxetine, which has repeatedly been reported in aquatic ecosystems. Worryingly, the primary target molecule of fluoxetine is conserved across a wide range of non-target species. As a result, by directly acting on the central nervous system and neuroendocrine pathways, fluoxetine can affect a range of ecologically important behavioural and physiological processes in wildlife. Despite this, the effects of environmentally relevant fluoxetine exposure on processes of sexual selection in aquatic biota remain uncertain. This is concerning as sexual selection processes directly influence mating outcomes and so are fundamental to individual fitness, as well as the viability of populations and species. To address this knowledge gap, we investigated the impact of 30-day exposure two environmentally realistic levels of fluoxetine (average measured concentrations: 30 and 380 ng/L) on a range of reproductive behaviours, as well as sperm quality, in the eastern mosquitofish (Gambusia holbrooki), a promiscuous freshwater fish with internal fertilisation. We focussed on these traits because reproductive behaviour and sperm quality are both crucial fitness determinants, and are known to be vulnerable to disruption by other chemical pollutant classes. We found that fluoxetine exposure impacts reproductive behaviour in fish at field-detected concentrations, altering both association time and copulatory behaviour (intromission frequency). Fluoxetine exposure, however, did not significantly impact sperm quality measures (i.e. performance and viability). In combination, our results indicate that fluoxetine exposure can alter reproductive behaviours with direct bearing on fitness in fish and, further, highlight the need for ecotoxicological testing using sub-lethal exposure concentrations and ecologically important behavioural endpoints.

TU325

Determining the effects of antidepressants on multiple behaviours in a marine and freshwater amphipod

S.A. Kohler, University of Portsmouth / Animal Physiological Ecology; A. Ford, University of Portsmouth / Biological Sciences; M.O. Parker, University of Portsmouth / Biological Sciences

Functional and physiological tests have been carried out by pharmacological discipline using anxiety-like behaviours including thigmotaxis (wall hugging) and scototaxis (light avoidance) on mice and zebrafish, using well-defined behavioural assays. These pharmacological methods have been translated to ecotoxicological studies on vertebrates but comparatively few have been done on invertebrate species. This ongoing study aims to translate these techniques to model crustaceans for the purpose of assessment of environmental risk using the antidepressant fluoxetine as a model compound. Specimens of the marine amphipod, Echinogammarus marinus and the freshwater amphipod Gammarus pulex were exposed to environmentally relevant concentrations of fluoxetine from 0.001-1 μg/L during 1 day, 1 week, and 2 weeks exposure. Activity was measured as swimming velocity and choice experiments were used to determine phototactic and thigmotactic response. Both E. marinus and G. pulex showed alterations in activity at concentrations as low as 1ng/L and as soon as 1 day compared to controls (P<0.05). Significant differences were observed in thigmotactic and phototactic behaviours both between treatments and with length of exposure. Results indicate that fluoxetine can have an effect on some amphipod behaviours at environmentally relevant concentrations. These results may have implications for future study design of these types of experiments and aid the development of high-throughput analysis on common laboratory invertebrate species.
Inter-species variability in the behaviour of a marine and freshwater amphipod

S.A. Kohler, University of Portsmouth / Animal Physiological Ecology; A. Ford, University of Portsmouth / Biological Sciences; M.O. Parker, University of Portsmouth

Inter-species variability in in standard ecotoxicology testing to assess environmental effects of contaminants. However, standardised invertebrate models are limited to relatively few species. Behavioural ecotoxicology is expanding with techniques and endpoints used in pharmacology being translated to other invertebrate and invertebrate species for use in ecotoxicology. Despite this, data on the control behaviours of model organisms such as crustaceans, and the inter-species variability in behaviours are currently under-studied. The aims of this study were to provide control data on a range of behaviours for use in ecotoxicological testing, using amphipods as model organisms. Behaviours commonly associated with anxiety in pharmacological studies including activity, phototaxis (light/dark preference) and thigmotaxis (wall hugging) were assessed in the marine amphipod *Echinogammarus marinus* and the freshwater amphipod *Gammarus pulex* using choice assays. Both organisms exhibited negative phototactic and positive thigmotactic behaviours (P<0.001 respectively) however, differences in sensitivity to these assays were observed between species. *E. marinus* showed a significantly greater sensitivity to the phototaxis assay than *G. pulex* (P=0.001), while the reverse was found for the thigmotaxis assay (P=0.001). Swimming velocity was used as a measure of activity. Significant differences were observed in swimming behaviours between species when exposed to a light stimulus (P=0.001) which may be attributed to differences in life histories between the two species. The results of this study provide evidence of phototactic and thigmotactic behaviours in two model crustacean species and describes two behavioural assays with potential for use in behavioural ecotoxicology. In this study we demonstrate that closely related species are capable of very different behavioural responses. The inter-species variability in sensitivity to behavioural assays found between the two amphipods in this study highlights the importance of control data on your model species for behavioural studies.

Physiological basis of individual tolerance to the benzodiazepine oxazepam in zebrafish (Danio rerio)

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Benzodiazepines are common contaminants in aquatic ecosystems. Among the most prescribed pharmaceuticals globally are the benzodiazepines (e.g. Valium), a class of psychoactive drugs used to treat anxiety and induce sedation. Benzodiazepines are persistent in the environment, and their target, the GABA-A receptor, is evolutionarily conserved throughout the vertebrates. Behavioural changes have been described for juvenile Eurasian perch (*Perca fluviatilis*) and Fathead minnows (*Pimephales promelas*); both species show increased swimming activity when exposed to benzodiazepines. Despite these findings, there is a lack of information on how individual variability in sensitivity to benzodiazepines may affect the swimming activity of zebrafish. Therefore, we investigated the effect of the benzodiazepine oxazepam on behaviour using the burbot, *Lota lota*. We found that high levels of oxazepam affected swimming activity, diurnal as well as nocturnal, while the environmentally relevant level had no detectable effect. There was also an effect on boldness, with fish exposed to high levels of oxazepam spending more time hiding than the control- and low level fish. Interestingly, the effects of high oxazepam were no longer detectible when the fish were tested again after being kept in water without drugs for five days. Our results suggest that effects of pharmaceuticals may be reversible, if the exposure duration is relatively short and the animal have the possibility to move to uncontaminated water.

Behavioural endpoints and biochemical biomarkers as tools to investigate effects of Citalopram in brown trout (Salmo trutta f. fario)

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Citalopram (CIT) is a selective serotonin reuptake inhibitor (SSRI) which is commonly used as an antidepressant. It binds to serotonin receptors for the serotonin reuptake from the synaptic cleft and thereby inhibits the reuptake of serotonin into the pre-synapse. Due to high consumption rates and moderate elimination during wastewater treatment, CIT is one of the most abundant SSRIs in surface water. Several studies showed that environmentally relevant concentrations of 1 µg/L may affect aquatic organisms. The aim of this study is to investigate effects of CIT in different life stages of brown trout (*Salmo trutta f. fario*) with focus on development, behaviour and individual health. Both, eggs of fish in the eyed ova stage and 8 months old juveniles were chronically exposed to four concentrations of CIT (1, 10, 100 and 1000 µg/L) in a in a semi-static three-block design accompanied by a control exposure. The larvae were exposed for 5 month at 7°C and 11°C, the experiments with juvenile fish were conducted for 4 weeks at 7°C. To investigate the effects of CIT on the embryonic development, mortality, hatching rate, and heartbeat rate were recorded. During the exposure, also behavioural endpoints were observed. Besides, several biomarkers indicative for fish health were investigated, such as cortisol-level, acetylcholinesterase activity, hsp70-level and the histological condition of the liver. After exposure to 1000 µg/L CIT, length and weight of both larvae and juveniles were significantly reduced. Furthermore, both groups showed an increased swimming activity and an increased swim up in the aquaria. In an artificial swimming measurement device (small aquaria with a diameter of 17 cm), videos were recorded for 20 minutes with the aim to quantify changes in the swimming behaviour. Due to the settings of the cameras, the aquaria were strongly illuminated and the water was not ventilated during the recording. This stress situation resulted in a high activity of fish except for those exposed to 1000 µg/L CIT. These moved significantly less with a lower velocity than the control fish. The results of both experiments make evident that 1000 µg/L CIT affects both larvae and juvenile brown trout, on one hand by making them more agile in the aquaria, but also by depressing stress-induced floury swimming. The study is embedded in the Effect-Net (effect network in water research) Project which is funded by the Wassennetzwerk Baden-Württemberg.

Assessing the direct and indirect effects of chemical contaminants on the behaviour, ecology and evolution of wildlife: A conceptual framework

K. Arnold, University of York / Environment; M. Saarisuo, Monash University / School of Biological Sciences; T. Brodin, Umea University / Department of Ecology and Environmental Science

Chemical contaminants, e.g. metals, pharmaceuticals, pesticides, are changing ecosystems via effects on wildlife. Most studies examine a limited range of endpoints, species and/or contaminants under laboratory conditions. Recent work explicitly based in ecological realism, however, reveals that chemical contaminants have complex impacts on wildlife and that these effects are strongly linked to life stage and environmental context. In this study we developed a conceptual framework that integrates direct and indirect effects of chemical contaminants on behaviour, under environmentally relevant concentrations and natural contexts. Within our framework, we show how the consequences of contaminants can extend beyond individuals. We use altered predator-prey interactions to demonstrate cascading contaminant effects through communities, exerting both positive and negative effects on distinct populations. Moreover, contaminants can be potent evolutionary forces selecting directly for compensatory behaviours or indirectly on downstream behaviours, via selection on resistance genes. To help implement our framework, we supply tools to design ecologically realistic experiments and risk-assessments. Although predicting effects of contaminants is complex, existing knowledge in ecology and evolution needs to be applied to this global environmental challenge.
multiple cues and both visual and chemical cues play an important role in choosing the most suitable mate. Therefore, it is crucial to understand if and how EDCs affect mate choice cues (e.g. visual and chemical cues), and further, if one cue is affected disproportionately. Accordingly, the aim of this study was to investigate the impacts of a 28-day exposure to 17α-ethinyl estradiol EE2 (measured concentration 12ngL−1) - a synthetic estrogen used in the contraceptive pill and a widespread contaminant of aquatic systems on visual and chemical communication in the guppy. To examine the impact of EE2 on mate mate choice, we ran a standard choice assay, which was conducted in two parts to disentangle visual cues from chemical cues. First, we allowed a single male (either control or EE2) to court two size-matched females (one control and one EE2-female). In this visual cue experiment, the male was only able to see the females, but not to smell them. Second, we introduced chemical cues (control and EE2-female) to the trial tank paired randomly with the females. We found that there was no significant effect of EE2-treatment on total time males spent associating with the females, when given only visual cues. There was, however, a significant effect on courtship ‘sigmoid’ display with both control and EE2-exposed males spending more time performing sigmoid displays for control females compared to EE2-exposed females. When males were presented with both visual (male and female) and chemical cues, no significant effects were observed. After exposure to 7 µg TBT L−1, eyes showed a darkening of the iris region after exposure to 4.5; 7 and 9 µg µTBT L−1. Histopathological changes and ImageJ/wrMTrack were compared. Exposure to undiluted samples. Exposure to serial dilutions of the effluent caused a significant effect of the effluent concentration on chemical cues, but also raises the possibility that EE2-exposed females that were paired with control chemical cues. Not only does our study uncover a previously unknown behavioural impact of EE2-exposure on chemical cues, but also raises the possibility that EE2-exposed males spending more time performing sigmoid displays for control females compared to EE2-exposed females. When males were presented with both female cues (control and EE2-female) to the trial tank paired randomly with the females. We found that there was no significant effect of EE2-treatment on total time males spent associating with the females, when given only visual cues. There was, however, a significant effect on courtship ‘sigmoid’ display with both control and EE2-exposed males spending more time performing sigmoid displays for control females compared to EE2-exposed females. When males were presented with both visual (male and female) and chemical cues, no significant effects were observed. After exposure to 7 µg TBT L−1, eyes showed a darkening of the iris region after exposure to 4.5; 7 and 9 µg µTBT L−1. Histopathological analysis of the retinal pigment epithelium (RPE) indicated a hyperpigmentation of the pigment epithelium villi and basal region in TBT exposed fish. In addition to these alterations, RPE invaginations, photoreceptor degeneration, iris epithelial cell atrophy and iris melanin condensation were observed. After exposure to 7 µg TBT L−1, swimming speed, swimming resistance, dark capture of Artemia nauplii and growth in weight were reduced by 85%; 60%, 33.6% and 56% relative to controls, respectively. Histopathological changes detected in the retina and iris may have reduced the fish visual exploration and prey detection capacity, which together with the detected effects in swimming endpoints might have led to a deficiency in prey capture and growth. These changes can reduce the chances of exposed fish to recruit to the adult population.

TU333 Chemosensory behavioral reactions of zebrafish larvae to environmental contaminants

SangHoon Jang, C.M. vom Berg, Eawag Swiss Federal Institute of Aquatic Science and Technology / Environmental Toxicology

Background: Selecting an appropriate behavioral response to a potential rewarding or threatening stimulus is critical for the survival of an animal. Therefore, organisms possess an innate ability to react to threatening and rewarding situations they are naturally exposed to. However, the ability to naturally respond to environmental cues can be affected by environmental factors and the behavioral outcomes become unpredictable. In particular, neuroactive and psychoactive substances in the aquatic environment released by wastewater treatment plants or agricultural run-offs might potentially change the perception and interpretation of natural cues by aquatic organisms, especially fish. Aim: We are investigating whether environmental contaminants lead to attractive or aversive responses in fish, and are additionally interested in the neuronal mechanism underlying the observed behavioral response. We aim to better understand how environmental contaminants change natural behavioral responses of fish in order to better predict their impact on the ecosystem. Methods: We are using zebrafish larvae as a model organism, because they are amenable for behavioral analysis and mechanistic dissection of complex processes. Larvae are exposed to a point source of test chemical at different concentrations and the behavior is tracked with an automated video recording system. Various parameters such as the larva’s space use, locomotor activity and velocity are evaluated. Active (toward) and passive (away from) responses are scored in an experimental arena. The arena is divided into three zones. Three responses are scored: (1) Avoidance responses ( avoidance avoided zone) and (2) attraction responses (attraction to zone). Results: We will dissect which chemosensory system and higher brain areas are involved in the behavioral reactions to environmental chemicals. This will advance our understanding of the impact of chemicals on fish behavior.

TU334 Urban sewage effluents into an alpine stream: are information on behavioural effects on Daphnia magna suitable to protect alpine cold adapted species? V. De Nisi, University of Milan - Bicocca (VAT IT12621570154) / Department of Earth and Environmental Sciences; V. Lenciioni, F. Bellamoli, MUSE-Museo delle Scienze / Dept of Chemistry and Biology; M. Barata, R.N. Alves, UFPE / Zoology; P.S. Carvalho, UFPE / Zoology - Universidade Federal de Pernambuco / Department of Zoology; K. Witte, University of Siegen / Department of Chemistry and Biology; E. Villa, EMA European Medicines Agency

Aim: We will identify which chemicals in WWTP effluents are among the most internationally used bioassays and for the toxicity of effluents. However, acute toxicity tests do not take into consideration endpoints that may provide early warning signals about the health of the exposed populations, such as changed or altered signal modulators. Altered behavioural signals could be induced at subthreshold concentrations which are significantly lower than the concentrations that cause mortality. By using WWTPs located in Alpine areas as surrogate of cold freshwater best adapted species. Mortality rate and behavioural responses (as swimming, analysed with two video tracking systems: Lollitrack Systems and Imager3WmTrick) were compared. For the behavioural investigations, exposure to undiluted samples. Exposure to serial dilutions of the effluent caused a significant effect of the effluent concentration on chemical cues, but also raises the possibility that EE2-exposed females that were paired with control chemical cues. Not only does our study uncover a previously unknown behavioural impact of EE2-exposure on chemical cues, but also raises the possibility that EE2-exposed males spending more time performing sigmoid displays for control females compared to EE2-exposed females. When males were presented with both visual (male and female) and chemical cues, no significant effects were observed. After exposure to 7 µg TBT L−1, eyes showed a darkening of the iris region after exposure to 4.5; 7 and 9 µg µTBT L−1. Histopathological analysis of the retinal pigment epithelium (RPE) indicated a hyperpigmentation of the pigment epithelium villi and basal region in TBT exposed fish. In addition to these alterations, RPE invaginations, photoreceptor degeneration, iris epithelial cell atrophy and iris melanin condensation were observed. After exposure to 7 µg TBT L−1, swimming speed, swimming resistance, dark capture of Artemia nauplii and growth in weight were reduced by 85%; 60%, 33.6% and 56% relative to controls, respectively. Histopathological changes detected in the retina and iris may have reduced the fish visual exploration and prey detection capacity, which together with the detected effects in swimming endpoints might have led to a deficiency in prey capture and growth. These changes can reduce the chances of exposed fish to recruit to the adult population.

TU335 Do silver and titanium dioxide nanoparticles influence the fish kainomone induced anti-predator defence in Daphnia magna? A. Beasley, University of Siegen; S. Hartmann, University of Siegen, Institute of Biology / Department of Chemistry and Biology; K. Witte, University of Siegen / Department of Chemistry and Biology

Daphnia possess a trait of phenotypic plasticity, whereby kairomones from fish induce growth and the formation of structures such as a spine or helmet. The resulting increase in body size, allows the daphnids to defend themselves from the predators in their natural environment. As the common link between green algae and fish in the food chain, daphnia are considered a key component in the freshwater ecosystems. According to K. Witt (1999), the main role of daphnia is that they may be necessary to prevent an ecological imbalance in the freshwater environment. Ag and TiO2 manufactured nanomaterials (MMNs) are widely used in the commercial industry because of their unique properties. Silver is known for its antimicrobial properties and is therefore used in soaps and bandages as well as clothing and washing machines. Titanium on the other hand is used in products such as sunscreen, paint and toothpaste because of the bright white pigment it contains. Due to their small size, nanoparticles are not being effectively removed from wastewater treatment plants and end up in freshwater systems such as rivers and streams. Filter feeders, like the cladoceran Daphnia, take up these nanoparticles and

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are therefore of particular scientific interest, to establish what impact the MNMs are having on the freshwater cycle and food-chain. In our study, we investigate the effects of Ag (NM300K) and TiO$_2$ (NM105) MNMs on the predator defence response; by chronically exposing Daphnia magna to fish kairomones and a range of nanoparticle concentrations. This experiment was conducted in accordance with OECD guideline No. 211 over a course of 21 days. For statistical analysis, we measured the body length, body width and tail spine length after each moult and calculated the nonmonotonic response of the reproductive parameters of the D. magna. A total of 15 daphnids is taken of each daphnid at the end of the experiment, to observe and compare the microscopic details of the anti-predator defences in the treated and untreated daphnia.

**TU336 Behavioral and Physiological Responses of Daphnia magna to Fluoxetine and Propranolol Exposure**

M.E. Nielsen, P. Roslev, Aalborg University / Biology and Environmental Science

Fluoxetine and propranolol are neuroactive human pharmaceuticals that occur as pollutants in surface waters. The potential effect of such pharmaceuticals on aquatic organisms including invertebrates has raised some concern but many adverse effects are not well characterized. In this study, 6 behavioral and physiological parameters in the freshwater Cladoceran Daphnia magna were compared for their responses to fluoxetine and propranolol exposure: mobility (dichotomous response), active swimming time, swimming distance, swimming velocity, swimming acceleration speed, and survival in the absence of food (starvation/survival). Changes in swimming behavior of D. magna were quantified by video tracking of single organisms followed by image analyses. Active swimming time and swimming distance appeared to be more responsive behavioral endpoints than swimming velocity and swimming acceleration. The EC50s for fluoxetine and propranolol determined from swimming time and swimming distance were comparable (1-2 µM). At low sublethal exposure concentrations (µg/L), nonmonotonic responses in swimming behavior were observed in D. magna. Behavior profiling estimated from multiple behavioral parameters showed that fluoxetine and propranolol stimulate swimming activity at 1-10 µg/L. EC50 values for fluoxetine and propranolol estimated from survival time in the absence of food (starvation/survival) were much lower than EC50 values estimated from changes in swimming behavior. Starvation-survival is strongly affected by energy metabolism and we suggest that this parameter can be a potential sensitive endpoint for determining adverse effects of pharmaceuticals to D. magna. Combining behavioral and physiological responses to high and very low exposure concentrations should be considered in models predicting adverse effects of pharmaceuticals to non-target organisms.

**TU337 How toxic is a non-toxic nanomaterial: Behaviour as an indicator of effect in Danio rerio (zebrafish) exposed to nanogold**

T. Bohu, North-West University / School of Biological Science; S. Brand, North-West University; V. Wepener, North-West University - School of Biological Sciences

Gold nanoparticles are used as drug delivery vectors based on the assumption that they have a low toxicity. Literature has however showed conflicting results over the last few years. This study aimed at investigating the toxicological effects of nanogold (nAu) over a range of indicators from sub cellular to whole organism level. Gene regulation, changes in oxidative stress biomarkers and swimming performance were assessed in Danio rerio (zebrafish) following exposures to nAu. Adult zebrafish were exposed to nAu for 96 hours, swimming performance was measured post exposure. Liver tissue was collected for DNA microarray and Real Time Polymerase Chain Reactions (RT-PCR) analyses to determine changes in gene expression (catalase, superoxide dismutase and metallothioneins). Whole body samples were stored in respective buffers for protein biomarker analysis (catalase, superoxide dismutase, acetylcholine esterase, malondialdehyde, cellular energy allocation and metallothionein). Swimming behaviour was assessed in 1.1 L Tecniplast™ tanks for a period of six hours and videos were analysed using Noldus EthoVision software. The critical swimming speed was performed in a Loloige® swimming tunnel, briefly fish were acclimatized within the chamber for one hour and then at a starting speed of 2 b/s with a 0.5 b/s speed interval, fish were swam until they were unable to keep up with the increasing water flow. The DNA microarray revealed that 20 mg/L was the least related to the control group. At 20 mg/L there was a significant increase in gene expression for all genes analysed but protein biomarkers showed no significant response. The behaviour results showed significant changes in distance moved, swimming speed, acceleration bouts, zone alterations and time spent within the top zone- responses which are seen in fish responding to toxicological stress. The exposed fish has a significantly lower critical swimming speed when compared to the control. Since swimming performance and social interaction during swimming is essential to life whole organism behaviour shows a toxicological response to nAu which is in agreement with genetic responses seen.

**TU338 The effects of silver and silver nanoparticles via different routes of exposure on behaviour in marine amphipods**

M. Vannucci-Silva, UNICAMP / Institute of Biology; S.A. Kohler, University of Portsmouth / Animal Physiological Ecology; G. Umbuzeiro, School of Technology, UNICAMP / LAEG; A. Ford, University of Portsmouth / Biological Sciences

Behavioural responses are an important endpoint because they provide a link between biochemical and ecological effects of environmental contaminants. Silver is increasingly being used in nanomaterials and, consequently, being released into the environment in different forms. The behavioural consequences of metal exposure in crustaceans have not been studied. Therefore, the aim of this work was to evaluate the effects on the swimming behaviour of the marine amphipod Echinogammarus marinus after exposure to silver, in its salt (AgCl and AgNO$_3$) and nanoparticle (E. marinus (n=20 per treatment) were exposed individually. The exposure via water was performed with Ag (from AgNO$_3$) for 5, 25 and 100 µg/L for 96 hours. The Ag exposure via food was performed with AgCl or AgNP (approximately 200 mg kg$^{-1}$) during 7, 14 and 28 days. The movements of the amphipods were tracked using a DanioVision™ system with EthoVision®XT software for behavioural analysis under 3 minutes dark/3 minutes light cycle. Differences in velocity of swimming, response to light and thigmotaxis were evaluated. In the Ag exposure via water, light significantly increased maximum velocity for all treatments (p<0.05) indicating an escape-related behaviour; excepting at 100 µg L$^{-1}$, where the maximum velocity had no difference between lights off and on (p=0.10), showing that silver at this concentration had a significant effect on the response to light; no significant effects were observed in frequency in centre zone for all treatments (p>0.05), although, cumulative duration in centre zone was significantly different when exposed to AgNO$_3$ 25 and 100 µg/L, where animals spent more time in the centre zone when was dark. Results from the exposure via food are currently undergoing analysis. The current results indicate that silver has effects on swimming and response to light behaviours in E. marinus, indicating that exposed animals in the environment could be more vulnerable to predation.

**TU339 Developing methods to determine aquatic invertebrate behavioural endpoints for regulatory ecotoxicology studies**


Under current plant protection product (PPP) regulation, Tier I aquatic ecotoxicology studies measure mortality (acute) or reproduction (chronic). For invertebrates, these parameters are used to address the current protection goals of maintaining populations. Agreed guidelines for reproduction studies for aquatic invertebrates are currently under development for 25 µg L$^{-1}$ E. marinus, however, for some substances, other taxa/species may potentially be more sensitive e.g. Ephemeroptera, Plecoptera, Trichoptera (EPT) species. Currently, there are no agreed methods for measuring reproduction endpoints for non-standard test species, such as EPT species. EPT species often live as larvae for a prolonged period and have an aerial adult stage, and thus assessing reproductive endpoints from a full life cycle may not be possible. Therefore, if an accurate long-term assessment of reproductive effects is desirable. Although behavioural endpoints (e.g. predator response, locomotion, feeding activity) do not directly relate to the protection goals of maintaining populations, they may still have a useful role in regulatory risk assessment. For example, for organophosphates (OPs) with steep dose-response curves and thus a narrow exposure window between acute and chronic effects, significant differences on reproduction can be due to mortality of adults rather than true reproductive effects; therefore, designing specific reproduction studies for e.g. EPT species may not be necessary for such substances. Instead, if a risk assessment were undertaken using acute and behavioural endpoints and acceptable risks were concluded, then it would be unlikely for effects at the population level to arise. Here we describe our experimental approaches to investigating invertebrate behavioural endpoints in one standard (Daphnia magna) and two non-standard (EPT: mayfly, caddis) test species that are suitable for use in regulatory toxicity tests, integrating the regulatory needs with the practicalities of ecotoxicology testing.

**TU340 The effects of sublethal doses of pollutants on crop pest, Spodoptera littoralis**

D. SIAUSST; Institute of Ecology and Environmental Sciences / Institute of Ecology and Environmental Sciences

Pesticides have long been used as the main solution to limit agricultural pests but their widespread use resulted in chronic or diffuse environmental pollution, development of insect resistances and biodiversity reduction. The effects of low residual doses of these chemical products on organisms that affect both targeted species (crop pests) but also beneficial insects became a major concern, particularly because low doses of pesticides can induce various effects. In addition to the negative effects, some studies highlighted unexpected positive - also called
hormetic - effects on insects, leading to surges in pest population growth at greater rate than what would have been observed without pesticide application. The present study aimed to examine the effects of sublethal doses of various representative products of large pesticide families used against a major pest insect, the cotton leafworm Spodoptera littoralis, and known to present a residual activity and persistence in the environment. Using an integrated approach from genes to behaviour, with the focus on the olfactory system and the sexual or feeding behavior of our crop pest model following application of sublethal doses of deltamethrin, methoxylin and chlorpyrifos. Whereas sublethal doses of methoxylin appeared to disrupt the feeding behavior of larvae, we demonstrated a hormeric response of males to sublethal dose of deltamethrin. We completed our study by molecular (qPCR), biochemical (proteomic, AChE activity and metabolic) and electrophysiological approaches in order to decrypt the involved mechanism in pesticide response as well as in the behavioral disruption.

TU341
The effect of copper nanoparticles on olfaction in rainbow trout (Oncorhynchus mykiss)
P. Borregaard, University of Lethbridge / Department of Biological Sciences; E. Mohaddes, University of Lethbridge; E. Lari, G.G. Pyle, University of Lethbridge / Biological Sciences
Fish rely on olfaction for their survival, growth, and reproduction. Impairment of olfactory function can pose a threat to fish survival on the small scale and population loss on the larger scale. Metal contaminants (e.g. copper) can impair fish olfactory function. Although the copper concentration in the water is not a direct threat to the fish, the effect of copper nanoparticles (CuNPs) on fish olfactory systems has not been well determined. The objective of this study was to investigate time-dependent effects of CuNPs and Cu" on olfactory acuity and olfactory-mediated behaviours of rainbow trout. To establish CuNPs or Cu" induced olfactory-impairment thresholds, inhibitory concentration (IC) curves were determined. Fish were exposed to a geometric dilution series of CuNPs or Cu" for 24 hours, and fish olfactory acuity was measured using electro-olfactography (EOG). Afterwards, fish were exposed to CuNPs or Cu" at concentrations known to impair olfaction by 50% (322 and 6.8 µg/L for CuNPs and Cu", respectively) for a 24 h or 96 h exposure period. The response of fish to a social cue (taurhoelic acid) was studied using EOG and a choice maze behavioural assay. After the behavioural experiment, fish olfactory rotisserie were dissected to investigate if there was any DNA fragmentation as a marker of apoptosis that might be induced by CuNPs or Cu". Results of EOG revealed that while a 96 h exposure to CuNPs caused a significantly greater impairment of fish olfactory function relative to a 24 h exposure to the same concentration, fish olfactory acuity partially recovered after 96 h of exposure. Continuous CuNPs exposure to the social cue supported the results of neurophysiological experiments. Although fish exposed to control water or Cu" for 96 h had an avoidance response to an alarm cue, those exposed to the CuNPs did not respond to the alarm cue. Results of DNA fragmentation indicated apoptosis was not the mechanism of olfactory toxicity for CuNPs or Cu" in the exposed fish. In summary, over the same exposure periods, CuNPs and Cu" differentially affected olfactory recovery. However, the mechanism of olfactory toxicity caused by CuNPs is not clear and needs further investigation.

Informed substitution of hazardous chemicals for circular economy: science and practice (P)

TU342
Perfluoroalkyl acids concentrations in liquid wastes: a survey campaign and implications for waste disposal
M. Peruzzo, Eurolab S. Valsecchi, University of Lethbridge / Department of Biological Sciences
A survey campaign has been carried out to determine the concentrations of twelve perfluorooalkyl acids (PFAA) in liquid wastes, before the disposal in dumpsite or incinerator. The common characteristic of these wastes was that they were classified as "wastes without dangerous substances" and could be disposed without specific treatments. Waste samples (about 120) came from solid waste treatment plants, drinking water and isolated sewage treatment plants, landfill leachate, solid waste storage facilities, car washing, septic tanks, laundry sludge and wastes from various industrial plants such as paper, food, wood, furniture, glass and pharmaceutical industries. The large spectrum of activities allows us to get a picture of the main uses of fluorinated compounds in the economic sectors and to evaluate the impact of the different sources. The percentage of samples which presented total PFAA concentrations greater than 1 µg/L was 65%. The maximum concentration measured was 900 µg/L. As regards the single congeners, the percentage of positive samples (i.e. > 0.05 µg/L) ranged from 5% to 37.5% for the different compounds. It is very interesting to note that PFOA and PFOS were found only in 5% of the samples, while the highest findings were for PFBA (57%) and PFBS (37.5%), highlighting the increasing diffusion of short chain PFAs respect to the already restricted C8-PFAs. It is also interesting to note that one of the samples with the highest concentrations was a found in the pharmaceutical industry, and it was an aqueous washing solution of water lignors. The overall survey underlines the need for a more accurate characterisation of wastes and the risk of transferring PFAA pollution from production sites to disposal sites, which can be located also in no-impacted areas.

TU343
Regenerated Textile raw materials: chemical contamination for LCA A. Franchi, Buzzi Laboratorio Analisi
It’s essential, for every actor involved in the supply chain of a textile company, to increase awareness that a regenerated material requires proper and specific evaluation standards. These should ensure compliance with private protocols and mandatory laws and also allow the proper control of the final product. CID (Italian Consortium for Detox Implementation) with the support of local actors (chemical laboratories) made a study concerning chemical contamination of regenerated materials in order to propose a PRSL (Product Restricted Substance List) for regenerated and recycled textile materials. The adoption of a PRSL for regenerated textiles would guarantee the safe re-use of these materials as an alternative to their disposal. This case study takes into account regenerated wollen textiles (high wool content > 70%) derived by post-consumables (knitted apparel, apparel made up by carded woven and combed woven), pre-consumables (combed and carded woven, knitted fabrics, spinning and twisting trimmings), and regenerated cotton-type materials (derived from denim recycling). Operational plan involved quantitative and qualitative assessment concerning the regenerated wollen type raw material used by carded spinning companies in Prato textile district, sampling (more than 100), raw type regenerated raw material selected by origin and type and more than 40 cotton-type regenerated materials) and chemical analysis (made by Buzzi Lab) of some priority groups of concerned substances: APEOS (Ethoxyiated Alkylphenols), Aromatic amines from azo-colorants, Chlorophenols, PFC (per-fluorinated compounds), Allergenic and Carcinogenic Dyes, Heavy metals. Analysis showed that about 150 sample were analysed and chemical contamination were found for aromatic amines (16% of total samples), APEOS (100% of samples), Chlorophenols (26% of total samples), PFC (62% of total samples) heavy metals (82% of total samples), Allergenic and carcinogenic dyes (6% of total samples). Data analysis permits to establish a PRSL (Product Restricted Substance List) protocol for regenerated raw materials with the aim to have an unique PRSL available for brands, manufacturing companies and every actor involved in the textile supply chain. The PRSL adption could improve the recycling of textile materials as an alternative to their disposal.

TU344
Challenges for a comparative risk assessment among conventional hazardous substances and alternatives for textile finishing, Two case studies: flame retardants and durable water and oil repellents, N. Fuentes, J. Damasio, V. Gonzalez-Andres, M. Diaz-Oritz, G. Janer, Leitat Technological Center
Some chemicals used in textile finishing processes are known to be toxic, persistent and bioaccumulative. Indeed, some of them (e.g. PFOA and DecaBDE) are listed in the Authorisation list and will be restricted after the sunset date. Alternative products are currently proposed by chemical companies for textile applications. Here we present two case studies (FLAREX and MIDWOR projects) focusing on the substitution of hazardous substances used as Flame Retardants (FRs) and Durable Water and Oil Repellents (DWORs). FRs are added to fabrics to inhibit the combustion process, and typically are products with a chemistry based on halogenates. DWORs are added to fabrics to repel water, oil and dirt, and typically are products with a chemistry based on long chain fluorocarbon polymers. These projects aim to support industry in the selection of alternatives. Alternative finishing additives available on the market were selected for laboratory validation of technical performance and industrial demonstration. In addition, a comparative risk assessment of conventional and these alternative formulations should be provided to ensure the reduction of environmental and human health impacts. One of the main challenges for a comparative risk assessment of these products is that the active substances in most of them are polymers and therefore are not subject to registration under REACH regulation. The potential human health and environmental impacts of these materials are related to their content or possible release of monomers (both with limited information). Moreover, the commercial formulations offered by chemical companies do not provide detailed composition. In fact, in the commercial products evaluated the chemical identity of the active substance was not reported in the safety data sheet. This is due to the lack of obligation to report inground metals from ash treating the hazardous classification of the mixture. Under this scenario, we propose to base the comparative risk assessment on the toxicological profile of the chemical family of corresponding monomers (based on the information supplied by the providers), and the operational conditions necessary for the application of each of the products (assuming that the risk mitigation measures will not change within an industrial setting). The results will support industry to select functional and safer alternatives.

TU345
Substitution of firefighting foams containing per- and polyfluorinated alkyl
substances (PFASs) per-and polyfluorinated alkyl substances (PFASs) are heat resistant and show a low flick resistance. Because of these properties PFASs are for example widely used in aqueous film forming firefighting foams (AFFF). PFASs are not degradable in the environment. Their release of AFFF into the environment causes a complex mixture of PFASs (such as PFOS and PFOA) need years to leach from top soil layers into the groundwater or into surface water. Short chain PFASs however reach ground water resources much faster due to their mobility in soil. Those contaminations already caused closed drinking water wells. Remediation is costly and long lasting. Although fluorne free foams are available and used at several European airports many firefighters hesitate to use them instead of AFFF. Restriction and authorization are regulatory measures under REACH which can be used to minimise releases of PFASs into the environment. An international regulation via the Stockholm Convention is possible as well. In addition the dialogue with stakeholders can lead to voluntary actions and may be an alternative measure to reduce environmental releases. Scientists and manufacturers need to be encouraged to develop environmentally friendly firefighting agents without fluorinated chemicals. Moreover, scientists, authorities and NGOs need to bring together knowledge about the new substances, such as analytical methods, and information on their fate and behaviour in the environment. This presentations provides an overview on regulatory actions regarding PFASs in the EU and further ideas how to substitute firefighting foams containing PFASs.

TU346
The Paradigm of Substitution - expand your view
M. Zimmer, ZVO e.V.; M. Metzner, Fraunhofer Gesellschaft

Many people mention substitution as the most promising option for risk reduction in the field of SVHCs. But it has to be considered that technical solutions are embedded in complex structure-effect relationships along of equally complexity. Many different properties and outcomes have to be considered. Otherwise a thorough assessment of the applicability of an assumes alternative will fail. The surface treatment sector as a major cross-sectional community of service providers has long term experience with innovation and substitution suggested by different players for many different reasons – risk reduction being one of them. In particular the SMEs have constantly been confronted with lots of different ideas and approaches. Hence they have deep insight into unexpected side effects and regrettable outcomes. And they are able to give indicators for real promising and applicable approaches to substitution. The authors will present some significant examples of substitution attempts – and they discuss arguments why they might be considered successful – or not.

TU347
A pilot case on how Socio-Economic Evidence can inform Risk Management decision making to assess Substitution versus Recycling for non-ferrous metals slags in safe use applications
H. Waertenschoot, M. vander Staeten, Eurometaux

The implementation of the European Circular Economy policy leads to more recycling, including closing the loop on substances. This policy combined with the increasing complexity of articles leads to increasing amounts of hazardous substances and impurities being available for recycling or reuse. Recycling processes in the metals sector produce besides pure substances for safe reuse, also final slags that collect some of the impurities that cannot be recovered at economical conditions. In parallel, the human health and environmental effects data generated by EU REACH and CLP Regulations lead to increasing hazard identifications and harmonised classification standards. Hazard endpoints of Very High Concern like CMR (carcinogens, mutagens and reproductive toxicity) or respiratory sensitisation may trigger substitution-based Risk Management Measures but also reduce the reuse in safe applications for “precautionary reasons”. In such cases, socio-economic evidence may be helpful to assess costs and benefits from a broader perspective, including Circular Economy and carbon footprint considerations.

A pilot study conducted at a non-ferrous plant specialised in the recycling of complex end-of-life articles and materials allowed to evaluate this impact and develop a tool for assessors to check how to what extent a change in a relevant hazard classification could impact the reuse capacity of final slags. The tool allows metal companies to assess their situation in respect to Substitution (materials loss or disposal) versus Reuse in safe applications.

Developments in the ecological and human health risk assessment of biopesticides: microorganisms, semiochemicals and botanicals (P)

TU348
Ecotoxicity of the hydrolate byproduct of three biopesticides on the unicellular green algae Chlamydomonas reinhardtii
D. Ballestero, J. Val, E. Lana, San Jorge University; E. Navarro, CSIC - Spanish National Research Council / Dept. Recursos marinos renovables; M. Piñeiro, Jorge University / Facultad ciencias de la salud; A.M. Mainar, Universidad de Zaragoza

Synthetic pesticides have been widely used in intensive production systems throughout most of the 20th century. However in the last decades, environmental and human health concerns demand safer substances, so research on biopesticides has been increased. Although a large number of studies have been published focusing on the biological activity of biopesticides on target organisms, studies regarding toxicological effects on non-target organisms, are scarce. The BIOCROP Project (Biopesticide development by chemical and biotechnological tools) has allowed the screening of several plant extracts for bio-activity against a selected set of crop pests and arthropod vectors. Some of these compounds have showed its effective value as biopesticides. The extracts will be optimized by means of traditional and supercritical CO2 technologies, as well as microbiological transformations. In the extraction process the organic and the aqueous fraction (hydrolates) have been separated and some showed active compounds, being capable to act as biopesticides. In order to exclude a negative effect on the environment, these products should be studied on non-target organisms. The aim of this study was to measure the acute ecotoxicity of hydrolates obtained of the semi industrial vapor-pressure essential oil extraction of three selected aromatic plant species; a domesticated Artemisia absinthium (Tereul, Spain), Dittrichia graveolens (Ciudad Real, Spain) and an experimentally pre-domesticated Lavandula lauiisieri (Toledo, Spain) using the algae Chlamydomonas reinhardtii as aquatic model organism. Results indicate that all of three extracts having biopesticide activity are likely to cause toxic effects on the photosynthesis of Chlamydomonas reinhardtii, being Lavandula lauisieri the most toxic compound followed by Artemisia absinthium with a very similar toxicity and Dittrichia graveolens. Consequently, the aim of this study is to evaluate the acute ecotoxicity of the hydrolate obtained from Satureja montana (Ejea, Aragón) by the traditional method of steam distillation on two organisms widely used as indicators of ecotoxicity: the freshwater crustacean Daphnia magna and the marine bacterium Vibrio fischeri. Both tests are standardized for the purpose of determining the toxicity expressed as EC50. Our results indicate that the hydrolate of Satureja is likely to cause toxic effects on D. Magna and V. Fischeri but only high dilutions (LC50 values in the range of 0,5% in both cases). These results allow us to know the possible environmental effect that these promising plant extracts can cause as a source of cosmetic and pharmaceutical applications with the aim of ensuring more environment-friendly processes and products.

Acknowledgements: We thank J. Burillo, for his generous cession of the extracts used in this study and the financial support of MINECO-FEDER (CTQ2015-64049-C3-2-R)

TU349
Ecotoxicological evaluation of the hydrolate byproduct of Satureja montana on Daphnia magna and Vibrio fischeri
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The increasing demand of natural products for cosmetic use, food or phytoterapy is based on the awareness about adverse effects on health and the environment. In particular, this work is focused on the plant Satureja montana (Lamiaceae), which has demonstrated a wide range of applications due to its important antimicrobial activity. Furthermore, plant metabolites of species have a sweet flavor and simple cultivation characteristics. The main components, thymol and carvacrol (oxygenated monoterpenes), are supposed to be responsible for these biological activities. Although there are a substantial number of studies where Satureja especies are evaluated for their biological and pharmacological activities, as well as its chemical characterization, limited data are available on ecotoxicological characterization. Consequently, the aim of this study is to evaluate the acute ecotoxicity of the hydrolate obtained from Satureja montana (Ejea, Aragón) by the traditional method of steam distillation on two organisms widely used as indicators of ecotoxicity: the freshwater crustacean Daphnia magna and the marine bacterium Vibrio fischeri. Both tests are standardized for the purpose of determining the toxicity expressed as EC50. Our results indicate that the hydrolate of Satureja is likely to cause toxic effects on D. Magna and V. Fischeri but only high dilutions (LC50 values in the range of 0,5% in both cases). These results allow us to know the possible environmental effect that these promising plant extracts can cause as a source of cosmetic and pharmaceutical applications with the aim of ensuring more environment-friendly processes and products.

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TU350
The Impact of the Hydrolate Byproduct of Three Biopesticides on the Soil Environment
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The extended use of synthetic pesticides has resulted, during the last century, in the pollution of the agricultural soil environments. As an alternative to these products, environmentally friendly biopesticides are, nowadays, being developed. Although biological activity of biopesticides on target organisms is well known, studies focusing on the effects on soil non-target organisms are scarce. The BIOCROP Project (Biopesticide development by chemical and biotechnological tools) focuses on the production and optimization of plant/fungal/agriwaste-based crop protectants via cultivation techniques, biotransformation, selective extraction and separations by traditional and supercritical CO2 technologies. In the traditional extraction process the organic and the aqueous fraction (hydrolates) have been separated. Both of them showed active components capable to act as biopesticides. In order to exclude a negative effect on the environment, these products were tested.
on soil non-target organisms (microbial community and earthworms). Soil microbial communities from an ecological farming crop have been exposed to three hydrocarbons, obtained by semi industrial vapor-pressure essential oil extraction, from three aromatic plant species: Artemisia absinthium, Ditrichia graveolens and Lavandula lauieteri. The effects on the microbial community has been assessed using the community-level physiological profile – CLPP-. This method relies on the ability of the microbial community for degrading different carbon sources present in the Biolog Ecoplates®. The acute toxicity of hydrocarbons was also tested by Eisenia fetida bioassays. Results indicate that hydrocarbons caused acute adverse effects in E. fetida, in particular D. graveolens and A. lauieteri (LC50 in the range of dilution of 10-2). All three biopesticides provoked changes in the soil microbial ability to degrade different carbon sources compared to control. These results allow for a better understanding of the impacts of natural crop protectants in the soil environment as a pest management alternative. Acknowledgements: We thank J. Burillo and J. Navarro for his generous cession of the extracts used in this study and the financial support of MINECO-FEDER (CTQ2015-64049-C3-2-R)

TU351

Acute toxicity of emulsifiable concentrate of Alpinia galangal essential oil against Cyprinus carpio

H. Kim, K. Kim, H. Jeon, Y. Choi, Y. Kim, S. Lee. Kyungpook National University Essential oils have exhibited their fumigational and topical toxicities on insect pests and they are developed as safe biopesticides. However, their use may cause potent toxic effects to non-target organisms in the environment. It needs to be determined the non-target effects (NTO) of non-target effects in the environment.

Alpinia galangal essential oil (AGEO) has been considered to control the outbreak insect pest, R. sin. sp. in South Korea. Acute toxicities of AGEO against Cyprinus carpio were assessed to understand its possible toxic effect on the representative aquatic organism. Cyprinus carpio in a static condition followed by OECD guideline 203 in 5 L beakers. As AGEOs were formulated for emulsifiable concentrates (EC) as an active ingredient, they were mixed with ethanol and tertigol in a ratio of 5:4:1. Tergitol is a surfactant and it did not show any toxic effect on the fish, so that it was used for the formulation of emulsifiable concentrate for AGEO.

Three different AGEOs were prepared according to their extraction methods as steam distillation, solvent extraction and supercritical fluid extraction. After the EC formulation was prepared, they were ready to expose to C. carpio to determine LC50 values. All emulsifiable concentrates of three different AGEO showed no mortality on the tested fish during 96-hour incubation. Therefore, 48-h LC50 values for the VFEO were under the toxicity criteria of level 3 for the pesticide to C. carpio standardized by Korea Rural Development Administration. Based on these results, AGEO can be considered to use as a natural insecticide.

TU352

Chronic toxicity of emulsifiable concentrate of cinnamon essential oils against Cyprinus carpio

H. Jeon, K. Kim, H. Kim, Y. Choi, S. Lee. Kyungpook National University

Recently, many researchers have developed natural insecticides to control insect pests. However, plant essential oils (EOs) are non-target organisms in the environment. Cinnamon EO is one of important EOs to be a potent candidate and is formulated as an emulsifiable concentrate (EC). As its use is recognized as safe, it needs to be determined its negative effect on the environment using bioindicators. To evaluate the negative effect on the ecosystem, chronic effects of cinnamon EOEC against Cyprinus carpio was determined in a static condition for 40 days. When cinnamon EOEC was added to the test solution, it showed acute and chronic toxicity to fish. To select an appropriate surfactants, 8 different types of surfactants (TWEEN 80, Sodium dodecyl sulfate (SDS), Nondient, Triton X-100, Sodium dodecyl Benzene Sulfonate (SDBS), Kolliphor, Tergitol and Mixture of SDBS and Nondient) were tested for the formulation and tertigol showed the lowest toxicity to the fish in an acute toxicity test. With the result of the acute toxicity of cinnamon EOEC, chronic toxicities of cinnamon EOEC was determined in five different concentrations for 40 days. Each concentration was triplicate exposed to 10 of C. carpio adults. The treated five concentrations were 0.08, 0.16, 0.46, 2.56, and 5.12 ppm and the mean survival rate was 8.48 ± 0.47. The survival rate of the control group was 9.33 ± 0.58 and the positive control containing ethanol and tertigol was 8.00 ± 1.00. The mean temperature and pH of the test water was 24.06 ± 0.58 °C and 7.51 ± 0.03, respectively. The mean of dissolved oxygen of the test water was 7.29 ± 0.07 mg/L and the mean of hardness was 82.14 ± 2.04 mg/L. After the complete exposure, the mean of length of alive fishes was 3.00 ± 0.17 cm and the weight was determined as 0.37 ± 0.17 g. With these results, cinnamon EOEC may be considered as safe, natural insecticides for the environment.

TU353

Thiosemicarbazone scaffold for the design of antifungal and antitoxiflaxotoxigenic agents: evaluation of ligands and related metal complexes

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Food safety is the safeguarding and protection of food from anything harmful affecting consumer health and is an extremely important issue facing the world. Food hazards can be divided into physical, chemical and biological. Examples of biological hazard are mycotoxins, that are toxic secondary metabolites produced by many species of filamentous fungi. Generally, mycotoxins represent a significant threat to human health as they can be carcinogenic, neurotoxic and toxic to endocrine or immune system. In particular, aflatoxins are a class of mycotoxin produced principally by two species of Aspergillus, A. flavus and A. parasiticus. Aflatoxins are found in various cereals, oil seeds, spices and nuts as a result of a fungal contamination that can occur in the field, during harvest, transport and storage. IARC has classified aflatoxins in Group 1 as carcinogenic agents to humans. The most dominant and potent aflatoxin is aflatoxin B1 and several studies indicate that high exposure to AFB1 can cause chronic toxicity and increases the incidence of hepatocellular carcinoma. A lot of methods can be applied to eliminate these toxins from food and guarantee the food safety and health concerns of consumers. Our research aims to develop new typologies of inhibitors of Aspergillus proliferation and of aflatoxin production, harmless to the environment and to human health. We have evaluated the biological activity of several thiosemicarbazone ligands starting from molecules of natural origin, like vanillin, perillaldehyde and their derivatives. In order to improve the biological activity, metal complexes were then synthesised. These molecules once synthesized and characterized, were initially tested to determine their antifungal and antitoxiflaxotoxigenic activity towards A. flavus. These compounds showed different efficacy in reducing fungal growth and mycotoxin accumulation. The most active compounds were used to perform cytotoxic and genotoxic tests on healthy human cells, particularly on human cell lines derived from the districts that can be exposed to xenobiotics. Furthermore, we performed toxic and genotoxic assays on bacteria and plant cells. In conclusion, this approach allows us to study the antifungal and antitoxiflaxotoxigenic activity of several thiosemicarbazones and to determine the potential risk for environment and human health with a view to use these compounds in field. Financial support: Fondazione Cariplo-Projekt N. 2014-0555, http://aflatox.unibas.it/

Understanding human and environmental exposure to chemicals in urban systems (P)

TU354

Electronic products are related with household exposures in Canadian residents

M.L. Diamond, C. Yang, University of Toronto / Department of Earth Sciences; L. Jantunen, Environment and Climate Change Canada; D. Tsirin, Cancer Care Ontario / Population Health and Prevention, Prevention and Cancer Control; L. Latifovic, Cancer Care Ontario; S. Harris, Cancer Care Ontario, University of Toronto / Population Health and Prevention, Prevention and Cancer Control Key Words: electronic products, hardware, household exposure, FRs and plasticizers Novel flame retardants (NFRs), polybrominated diphenyl ethers (PBDEs), organophosphate esters (OPEs), and phthalates esters (PAEs) have wide applications as flame retardants (FRs) or/and plasticizers in consumer products, building materials, and industrial uses. Their widespread use has led to pop-ulation-wide exposures (Carignan et al. 2017; Hammel et al., 2016; Hoffman et al. 2015). Some of these exposures have been related to adverse health effects (e.g., Carignan et al. 2017). Therefore, information on major exposure sources is needed to reduce exposures and ultimately prevent adverse health outcomes. Here we report on a household exposure study of Canadian women by determining levels of selected FRs and plasticizers in paired household air and dust, hand surface wipes of participants, as well as wipes of principal household electronic devices, including their cell phones. PAEs had the highest overall concentrations followed by OPEs by approximately one order of magnitude, and NBRFs and PBDEs (three orders of magnitude less than PAEs). Multiple compounds were found in wipes of individual electronic products suggesting either their usage in many products or migration into the surface polymer of these products from other sources indoors. Statistical analysis showed that PAEs profiles on a participant’s hands most frequently resembled the profiles found in that person’s hand products from other sources indoors.
plasticizers. Our results indicate that participants are exposed to various FRs and plasticizers through their daily household environment. The Canadian adults’ external exposure of hands to FRs and plasticizers were related to the levels in their household electronic products, particularly handheld devices such as cell phones. Handheld devices could contribute to human exposure through direct contact during use while large and stationary electronic products could be important sources and sinks in household environment.

TU355 Modelling diffuse emissions and fate of engineered nanoparticles used in outdoor paints to urban surface waters at high spatial and temporal resolution M.D. Núñez, University of York / Environment; A. Praetorius, University of Vienna / Environment Department & CESAM; A.L. Machado, University of Aveiro / CESAM

The expansion of the nanotechnology sector is leading to an increased use of products containing engineered nanoparticles (ENPs) in outdoor urban environments. Outdoor materials, such as construction materials, paints and coatings, are subject to weathering and ageing processes and will consequently lead to emissions of ENPs to the surrounding environment over time. Data on measured environmental exposure concentrations are still lacking for ENPs. Until analytical and monitoring techniques for ENPs in environmental matrices become available, modelling tools are the best approach to estimate exposure levels. Furthermore, models can analyze a wide range of potential scenarios and predict possible future trends of urban exposure which cannot be achieved by monitoring. In this study we propose a novel modelling approach that combines an emission and a fate model for ENPs with high spatial and temporal resolution for an urban environment. The model was applied to the study of titanium dioxide (TiO₂) ENP emissions when incorporated in outdoor paints in the city of York (UK). The model emission calculations are based on locally collected data on outdoor paint usage (outdoor paint application ratios and frequency of application for York) and information about the sewage network connectivity around the city. Reliable and official sources of information, such as Yorkshire Water and York City Council, and surface water characteristics acquired from an extensive and local monitoring campaign performed in the rivers Ouse and Foss, helped to parametrize the river fate model. Using the model, the transport and fate of TiO₂ ENPs in the rivers circulating within the city (the Ouse and the Foss) could be studied and spatially resolved results obtained. The identification of hot spots of emissions within the city and the study of ENP transport and fate are accomplished by this approach.

TU356 Occurrence and human exposure of parabens, triclosan and triclocarbon in personal care products from Korea S. Mok, Hanbat University / Marine Sciences and Convergent Technology; J. Lim, Hanyang University; M. Lim, Seoul National University / Environmental Health Sciences; J. Park, Seoul National University / Environmental Health Science; K. Lee, Seoul National University, Graduate School of Public Health; H. Moon, Hanyang University / Environment Department & CESAM; A.L. Machado, University of Aveiro / CESAM

Parabens (p-hydroxybenzoic acid esters), triclosan (TCS) and triclocarbon (TCC), have been extensively used in various cosmetics and personal care products (CPPCs) as preservatives due to their antimicrobial activities. However, little is known about the occurrence and exposure levels of parabens, TCS and TCC associated with the consumption of CPPCs in our daily life. In this study, ten parabens and their metabolites, TCS and TCC were measured in 243 CPPCs, which comprised of leave-on products (n=157), rinse-off products (n=59) and baby care products (n=31), collected from Korean market during 2016-2017, using liquid chromatography-tandem mass spectrometry (LC-MS/MS). Among ten parabens, methyl paraben (MeP) showed the highest detection rate (57%), followed by propyl paraben (PP, 49%) and butyl paraben (BuP, 41%). TCC had only 20% of detection rate and TCS was rarely detected in the samples. Total concentration of parabens widely varied with ranging from < LOQ to 10200 µg/g. Concentrations of TCS and TCC ranged from < LOQ to 340 ng/g and < LOQ to 14.0 ng/g, respectively. Higher concentrations of parabens (> 1000 µg/g) were found at skin cares, sunscreen, face cleanser, eye-liner, body/hand lotions and lipstick. The daily exposure levels of parabens and TCS and TCC were calculated using consumption rates and exposure factors, obtained from questionnaire-based survey and previous studies, and concentrations measured in our study. The mean daily exposure levels of parabens were 16.2 and 0.14 µg/kg body weight/day for mothers and their infants of Korea, respectively. Among CPPCs, some leave-on products such as skin cares, body/hand lotions, and sunscreens were the major contributors (> 80%) to total exposure levels of total parabens.

TU357 Characteristics of exposure factors for consumer products in Korean infant and caregivers pair K. Lee, Seoul National University, Graduate School of Public Health; M. Lim, Seoul National University / Environmental Health Sciences; J. Park, Seoul National University / Environmental Health Science

Concerns about potential health risks of chemicals in consumer products like cosmetic, personal care products, food containers are growing. Especially infant and children are more vulnerable to chemical exposure compared to adults. Since exposure to these chemicals could be determined by consumer products usage pattern, an accurate measurement of the usage patterns of consumer products is important for realistic exposure assessment. The aims of this study were to determine exposure factors of consumer products for child and mother and analyze the relationship between consumer exposures of caregiver and infant. We determined the exposure factors of 12 kinds of cosmetics (3 basic cosmetics, 1 UV protection products, 3 body products, 2 cleansing products) for adults and 10 kinds of consumer products (2 cosmetics, 3 oral supplies and 7 household products) for children and 11 kinds of food containers for household. Survey was conducted on 505 mother-infant pairs from Oct. to Dec. 2015 in Seoul metropolitan area by a structured questionnaire. The number of subjects were determined by proportioning sampling based on the frequency of use and the composition ratio in children’s sex and age distribution from 0 to 4. All cosmetics investigated in this study were used on a daily basis and usage rates from 52.1% to 98.0%, except 9.9% for hair styling product and 7.7% for deodorant. The frequency of food intake by food containers ranged 2.52 to 17.39 times a month. The use of children's oral supplies varies according to the age of the child. There were a significant difference in the mother’s usage rates of lotion, hair products and vinyl package food by age of children. These exposure factor characteristics could be useful input data for exposure risk and assessment for chemical regulation.

TU358 Analysis of metabolites of organophosphate and pyrethroid pesticides in urine from Italian children N. Bravo, CSIC-IDAEA / Department of Environmental Chemistry; J. Grimalt, Institute of Environmental Assessment and Water Research IDAEA CSIC / Department of Environmental Chemistry; B. Bocca, G. Calamandrei, A. Alimonti, Istituto Superiore di Sanità

Organophosphate (OP) and pyrethroid (PYR) pesticides are commonly used in agriculture, domestic environments and gardening. They eliminate insects because of their strong potential to disrupt the brain and nervous system of these organisms. Unfortunately, this neurotoxic effect is not selective enough as to avoid potential damage to other non-target species, including humans. OP and PYR pesticide exposure has been related to several human health effects, including respiratory, digestive, productive and neurological problems, among others. Children are more vulnerable than adults to environmental pollutant exposure because their organs and metabolism is still under development. Thus, their detoxification mechanisms are not yet mature. Once in the human body, OP and PYR pesticides are typically metabolized and excreted in urine within 4–48 hours after exposure, depending on the compound. Organophosphates are metabolized into dialkyl phosphates (DAPs) and hydroxyl phosphates (DHPs), such as diethylcarbamyl phosphorop (DEAMPY, metabolite of pirimiphos), 4-nitrophosphor (PNP, metabolite of parathion), malathion dicarboxylic acid (MDA, metabolite of malathion), 3-chloro-4-methyl-7-hydroxycoumarin (CMHC, metabolite of coumaphos), 2-isopropyl-6-methyl-4-pirimidyl (IMPY, metabolite of dialzinón) and 2-dethylinamino-6-methyl piriminid-4-ol (DEAMYP, metabolite of pirimiphos). Concerning pyrethroids, most compounds such as permethrin, cypermethrin, deltamethrin, allethrin, resmethrin and fenvalerate are metabolized into one single compound, 3-phenoxbenzoic acid (3-PBA). On the other hand, cyfluthrin is metabolized into 4-fluoro-3-phenoxbenzoic acid (4-F-3-PBA). Therefore, 3-PBA and 4-F-3-PBA can be used as a biomarkers of the most common PYR pesticides. The determination of the above mentioned compounds was performed using isotope dilution liquid chromatography-tandem mass spectrometry (LC-MS/MS). Human urine samples (n=199) from Italian children at 7 years were analyzed. Neuropsychological and psychomotor development of the children was assessed at 18–40 months by using the BAYLEY scales. In addition, neuropsychological development and intelligence were assessed at 7 years by using the NEPSY-II and the WISC, respectively. The compounds detected the most were DEAMYP (98%), PNP (97%), 3-PBA (91%) and TCPP (87%). The metabolite showing the highest concentration was DEAMYP with a median of 3.0 ng/mL followed by PNP, 3-PBA and TCPP with medians of 1.3, 0.56 and 0.47 ng/mL, respectively.

TU359 PAH levels in parturient and newborns from Aveiro region, Portugal, M. Monterio, Aveiro University / Biology; M. Fraga, Biology Department CESAM Aveiro University; C. Gravato, Faculdade Ciências da Universidade de Lisboa / department of Biology & CESAM; C.J. Silva, University of Aveiro / Biology Department & CESAM; A.L. Machado, University of Aveiro / CESAM Department of Biology; A.M. Soares, University of Aveiro / department of Biology & CESAM; S. Loureiro, Universidade de Aveiro / Biology

Environmental exposure to humans may be critical in some residential and working areas, and therefore biomarkers can and should be used as early warning tools to identify areas, and therefore biomarkers can and should be used as early warning tools to determine exposure factors of consumer products for child and mother and analyze the relationship between consumer exposures of caregiver and infant. We determined the exposure factors of 12 kinds of cosmetics (3 basic cosmetics, 1 UV protection products, 3 body products, 3 body products and 2 cleansing products) for adults and 10 kinds of consumer products (2 cosmetics, 3 oral supplies and 7 household products) for children and 11 kinds of food containers for household. Survey was conducted on 505 mother-infant pairs from Oct. to Dec. 2015 in Seoul metropolitan area by a structured questionnaire. The number of subjects were determined by proportioning sampling based on the frequency of use and the composition ratio in children’s sex and age distribution from 0 to 4. All cosmetics investigated in this study were used on a daily basis and usage rates from 52.1% to 98.0%, except 9.9% for hair styling product and 7.7% for deodorant. The frequency of food intake by food containers ranged 2.52 to 17.39 times a month. The use of children's oral supplies varies according to the age of the child. There were a significant difference in the mother’s usage rates of lotion, hair products and vinyl package food by age of children. These exposure factor characteristics could be useful input data for exposure risk and assessment for chemical regulation.

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particulate consent. Levels of PAHs equivalents were measured by a fluorescence methodology and were correlated with exposure to tobacco smoke as well as with other information regarding mother’s lifestyle (e.g. urban or rural residential area, exposure to vehicles exhaust). In general, the studied group presented higher PAHs levels in the placenta and lower PAHs levels in the umbilical cord blood. The low molecular weight PAHs (naphthalene and phenanthrene) measured in placenta presented higher concentration than high molecular weight PAHs (pyrene and benzo[al]pyrene). More increased levels of benzo[a]pyrene and phenanthrene equivalents were associated with exposure to vehicle exhaust, while higher levels of benzo[a]pyrene were associated with exposure to tobacco smoke at work. The highest naphthalene, pyrene and BaP equivalents levels were found in homogenized placenta of mothers who smoked in the third trimester of pregnancy. No significant correlations were found between PAHs levels and anthropometric data of newborns, but in general, higher PAHs levels were found in newborns with lower weight, head circumference, and length. Maternal-infant biomonitoring can be a major asset in evaluating environmental exposure to contaminants, which can also provide high value information for preventive medicine.

TU360
A modelling framework to link aggregate exposure pathways with internal exposures and potential bioactivity
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The aggregate exposure pathway (AEP) model is a conceptual framework to help all stakeholders link exposure information including (i) production, use and release, (ii) chemical fate and concentrations in various multimedia (urban and rural environments, biota), (iii) external exposures (e.g., contact rates), and (iv) internal exposures (e.g., blood concentrations) for human and ecological receptors. Some exposure models include elements of the AEP framework and are useful tools for organizing data, quantifying chemical concentrations throughout the source-to-exposure continuum and identifying research needs to address uncertainty in chemical evaluations. We present an overview of the Risk Assessment Identification And Ranking-Indoor and Consumer Exposure (RAIDAR-ICE) modelling framework. RAIDAR-ICE includes direct and indirect near-field exposures and can include far-field exposures for aggregate human exposure assessment. The RAIDAR-ICE model is parameterized in this case study for about 200 organic chemicals comprising a broad range of chemical properties representative of commercial chemicals to demonstrate model applications for exposure and risk-based prioritization. Based on assumed emissions to air in the indoor environment, intake fractions (used for ranking exposure potential) range from 0.0018 to 0.37 emphasising the relatively high potential for human exposures to certain chemicals from indoor environments. The exposure model calculated critical emission rates of the case study chemicals span approximately 8 orders of magnitude. Including estimates of actual chemical use, the model calculated critical emission rates can be used to gauge potential risks and provide guidance for proposed new chemical use. Using in vitro bioactivity data from the ToxCast program as an assumed “effect threshold”, the critical emission rates of the case study chemicals span approximately 8 orders of magnitude. Including estimates of actual chemical use rates allows for the calculation of risk (bioactivity)-based estimates; the results of which span at least 15 orders of magnitude. Recommendations for addressing uncertainty in the model and its required input parameters are presented.

TU361
ENVIRONMENTAL IMPACT OF LEAD MINING ON THE BIO-ECOSYSTEM IN ISHIAGU TOWN OF EBONYI STATE IN SOUTH-EASTERN NIGERIA
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Lead is a soft, ductile metal found naturally in the environment and accounting for 0.0016% of the earth’s crust. However, due to its ubiquitous nature, it is used in several industrial processes which can result to severe environmental pollution which can pass across food chains to animals and man. In 2010, about 400 deaths especially among children in Bukkyum and Anka LGA of Zamfara State, Nigeria, due to chronic lead toxicity were reported by Medecins Sans Frontieres (MSF, Holland) to the health authorities (UNEPO/CHA 2010). The cause of the high mortality was acute and chronic lead poisoning as a result of massive environmental contamination from artisanal mining and processing of gold in Pb-rich ore by poor headmen and farmers. This outbreak was reported as the worst in modern history (UNEPO/CHA 2010). Open-pit mining of lead in the Isehiagui Region of Ebonyi State since 1965 has exposed large volumes of marcasites, pyrites and tailings contaminating the environment and food chain pathways. The research was designed to investigate the environmental impact of lead mining on the bio-ecosystem of Ishiagiu town and environs. Soil, water, grasses/plants, food, fish and quarry dust were collected between March and May 2017, processed and analysed for lead concentrations. All water samples exceeded WHO recommended safety limits for lead. Soil and food samples contained values. Sampling of Ivo River, the main communal water source showed links to upstream pollution as the river passes through lead mining fields. The result showed the negative impact of lead mining in Ishiagiu and the need for regulatory agencies/government to take measures to avert consequences of lead poisoning in human beings.

TU362
Evaluation of potential risk of rare earth element contamination from leachate originating from electronic waste disposal
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Rare Earth Elements (REEs) form critical elements required in technological appliances. Their presence in electronic waste gives leads to environmental pollution. Therefore, there is a constant necessity for accurate data and reliable fast analytical methods. This review discusses and compares the methods given for the determination of rare earth elements and heavy metals in electronic waste and other associated environmental contamination. The methods are compared and discussed. This study further gives attention to the present methods and instrumental techniques with a focus on rare earth elements in electronic waste and its disposal into the environment. Key words: Rare earth elements; electronic waste; fusion optimisation; spectroscopy; wastewater

TU363
A stonewall snail as a new biomonitor of metal contamination in the urban environment
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Papilifera papillaris (O.F. Müller) is a small pulmonate gastropod commonly dwelling on stone walls and monuments in Italian and Mediterranean urban environments. This widespread, low-vagile and omnivorous organism, which barely interacts directly with soil and inhales fine particles, is a promising indicator of urban pollution and contamination. As a marine gastropod, it organisms widely used as biomonitor of urban pollution, i.e. mooses, lichins and vascular plants, accumulate particles of soil and rock dust, making it difficult to recognize the element contribution from atmospheric deposition and the metal bioavailability to consumers. By analysing the chemical composition of the shells, soft tissues and faeces of snails collected from vegetated walls, at roadside and community gardens, this three summer collections (Capannori, Lecco, Aosta) and five (Capannori, Lecco, Aosta) soft tissues of P. papillaris (purged of the gut contents) showed the highest Cd, Cu and Zn concentrations at the most trafficked sites. Data from faeces suggested that this species ingests large amounts of inorganic particles scraped from the stone surfaces, as indicated by the very high Al, Cr, Fe, Mn, and Pb concentrations. Most lithophilic elements and Pb are scarcely absorbed in the snail digestive tract and soft tissues mainly accumulate Cd and essential elements such as Cu, Zn and Mn.

Although the chemical composition of the shells was characterized by low concentrations of all analyzed elements, the samples collected at the most trafficked sites had significantly higher Cu, Fe, and Zn concentrations. This bioaccumulation in P. papillaris shells likely remains after death, potentially providing a historical record of the snail exposure to metals over lifetime.

TU364
Metals Distribution in Urban Garden Soils in Greater Victoria, BC, Canada
M. Dodd, Royal Roads University / School of Environment & Sustainability
This study was conducted to determine heavy metal distribution in surface soils in Greater Victoria, BC, Canada. Over 160 soil samples were collected from 190 residential and community gardens and analyzed using a portable XRF. A subset of the samples were analyzed by ICP-MS. A comparison of the XRF metal concentrations to the ICP-MS data indicated that the XRF was a suitable technique for the rapid analysis of the large number of samples collected. Elevated concentrations of metals including Pb, Cu, Cr and Zn, were found in some garden soils. Based on homeowner interviews, historical maps and archival reviews, Pb contamination was primarily attributed to the use of leaded paints and housing maintenance practices. Potential sources of the other metal contaminants included the use of wood preservatives, septic fields, automotive
repair and old orchards. An interactive map of metal distribution based on the data obtained was developed and made available to the public. Metal bioavailability was assessed using an in vitro bioaccessibility assay and the data used to assess the risk associated with soil ingestion. The estimated daily intake was determined for each element incorporating metal bioaccessibility data. Using the median concentrations, the calculated EDI values were well below the respective tolerable daily intake suggesting that the risk associated with ingestion of metal contaminants was minimal. Dandelion samples were also collected and analyzed as surrogate plants to determine potential metal uptake. Metal bioaccumulation factors and translocation factors for the dandelion samples also suggested that the potential for the uptake of the metals studied was low. However there were isolated gardens with elevated Pb concentrations which were identified as being of concern. Recommendations for limiting Pb exposure in these gardens were provided to the homeowners.

TU365
Soil quality analysis, a lever for identifying sources of trace elements and managing urban allotments for urban agriculture production
M. Letragor, C. Delnoye, A. Di Guardo, L. Rouan, Aghyle Unit
Projects seeking to produce home-consumption vegetables in urban areas and, more generally, to set up agricultural production inside cities, are increasingly numerous. Agglomerations and local public authorities also need references on thresholds of contamination in trace element and their transfer into plants. At European level, soil quality regulations are not homogeneous; in France, the legislation is mainly based on the contents in vegetables or fruits, the link with the soil never made. Some studies have highlighted the potential risk of metal contamination of vegetables grown in urban areas and the lack of site-specific risk assessments. However, experimental trials are still lacking on the potential of using urban soil as a good substrate for producing vegetables for domestic consumption. We assessed the quality of the soil on a site in the Rouen agglomeration (Normandy, France) for three partners: pasture, a former creation and mobile market gardening area. However, the city raises questions about the future management of this last area already cultivated for many years. We have analyzed the manure physicochemical characteristics of soils, the trace elements (Pb, Cu, Ni, Zn, Cd, Hg) in certain vegetables and fruits and in soils, as well as the history of agricultural practices since the sixties and the topographe of the site. Transfer coefficients in consumption vegetables have also been quantified. Our results showed that the zones had a physical and chemical heterogeneity due to the effects of the different cultivation techniques used, the urbanization and the topography of the site. Some metals contents are often above the recommended limits, and soil conditions (pH = 8) significantly reduce the mobility of metals. The concentration of Pb in some of the cultivated urban samples was above limits, which makes gardening practices unsuitable for the area. Our results demonstrate that site-specific studies are needed before planning urban cropping areas, and educating urban gardeners about sustainable cropping techniques is a priority for safe feeding.

TU366
Vertical movement of PCBs in agricultural soils impacted by an historical contaminated site: using SoilPlus model to predict discharge, dynamics of movement in soil, and rhizoremmediation potential
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Among the national priority polluted sites, the SIN Brescia Caffaro located in a mid size city Brescia, (200,000 inhabitants) in northern Italy. The site derived from the activity of the former Caffaro s.p.a., a chemical factory among the largest former polychlorinated biphenyls (PCBs) producer in Europe, which produced such chemicals for more than 50 years up to mid 80’s. About 100 Ha of agricultural areas were contaminated by a mix of Persistent Organic Pollutants (POPs, mostly PCBs, dioxins, furans) and heavy metals (Hg, As) in variable concentrations, often exceed the referred values. The contamination of runoff irrigation with contaminated waters. PCBs were measured in three different agricultural areas and in three different points per area, in vertical cores up to 1 m depth. The resulting samples were representative of 0-10, 10-20, 20-30, 30-40, 40-60, 60-80, 80-100 cm. The results of concentration measurements with depth (for about 80 PCB congeners) confirmed a general tendency of PCBs to be confined to the surface layers and not to vertically penetrate. PCB 28 ranged from 150 to 250 µg/kg in the top 30 cm to about 0.6 µg/kg at 1 m depth; PCB 209 (peculiar of Caffaro production) ranged from 1500 to 13000 µg/kg in the top 30 cm, descending to about 13 µg/kg at 1 m depth. A gradient was also observed along the runoff water flow direction. These concentrations of PCBs were then compared to those obtained by the SoilPlus model (a multilayered dynamic multimedia fugacity model) and used to predict discharge amounts and conditions regulating vertical movement. The objective was to reconstruct soil concentration profile during the historical contamination to predict discharged amount, potential for additional vertical movement, and conditions regulating chemical bioavailability for future PCB rhizoremidation.

TU367
Metals and metalloids in inhalable fractions of urban road dust
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Road dusts are highly enriched with metals and metalloids such as Cu, Zn, Pb, and Cd, due to road surface material and the wear of automotive components. Despite the importance of road dust as a source of inhalable particles (< 10 µm), little has been published regarding elemental enrichment patterns in particle sizes relevant to inhalation exposures. The goal of this study is to evaluate the contribution of road dust to airborne particulate matter, focussing on metals and metalloids in the inhalable particle size range. Road dust samples were collected from a variety of street types in 2015-2016 in collaboration with the City of Toronto, representing a total road length of about 840 km. Two types of samples were generated by the regenerative-air sweepers: the bulk hopper debris and finer dust box samples. The 50th percentile particle size diameter of the dust box samples was determined by laser analysis to be 9.4 µm, which represents the inhalable fraction. A total of 64 samples (32 inhalable and 32 bulk samples) were subjected to an acid digestion (HF, HClO4, HNO3 and HCl) followed by multi-element determination using Inductively-Coupled Plasma Mass Spectrometry (ICP-MS). Results showed that the inhalable fractions of road dust were enriched with metals and metalloids relative to the bulk debris, including Cd (0.55 vs. 0.25 µg/g), Zn (649 vs. 252 µg/g), Pb (8 vs. 2.2 µg/g) and Pb (80 vs. 8 µg/g). The enrichment of elements of known toxicity in the inhalable fraction is of particular concern, given the bioaccessibility of this particle size range. Available data on the total weight of road dust collected by the City of Toronto each year, combined with the elemental concentrations of the road dust determined in the present study, provides the means to calculate annual flux estimates. For example, Pb loadings in the inhalable fraction alone are estimated to range between 70 kg/yr and 141 kg/yr, which is a significant source relative to the city’s total (513 kg/yr) Pb emissions reported by industries and municipal facilities in the National Pollutant Release Inventory. This example demonstrates the importance of non-exhaust forms of traffic emissions which have not been included in emission inventories to date. Next steps will involve the collection of whole, unfractonated road dust samples from a variety of road types for particle size distribution analyses to refine elemental loadings in each size fraction.

TU368
Sequential extraction and particle size distribution of Cd, Cu, Pb and Zn in street dust of Belgrade (Serbia)
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The aim of this study was to investigate the differences in distribution of Cd, Cu, Pb, and Zn in mobile phases extracted from different size-fractions in street dust particles from Belgrade, the Capital of Serbia. The metals investigated were chosen as the most significant pollutants according the European Environment Agency. The street dust samples were collected in summer 2016 at three different locations. The samples were fractionized into three sizes with diameters of: < 63 µm, 63 – 250 µm, and 250 – 500 µm. From different size fractions the metals were isolated into three fractions using a modified sequential extraction procedure after Tessier (Tessier et al., 1979); adsorptive and ion-exchangeable phase (using ammonium acetate); moderately reducible phase (using ammonium oxalate and oxalic acid); and organic sulphide phase (using hydrogen peroxide acidified with nitric acid). These fractions were analysed by inductively coupled plasma optical emission spectrometry (ICP-OES) using an ICP iCAP6500 Duo-Thermo Scientific instrument. The results showed that the concentrations of the metals (based on the sum of these three fractions) were in the following order: Zn > Cu > Pb > Cd. Comparison with the Serbian national Regulations demonstrated that the concentrations of Cu and Zn in these three fractions were higher than the Maximum allowed values at some locations only, indicating serious contamination with these metals at some locations. The phase partitioning study revealed that Zn and Cu were bounded mainly in the second phase. Pb and Cd were predominantly associated with the inhalable fractions. In some samples predominance of Cd was associated with the third phase while Cd was in one sample predominantly associated with the first phase. Detailed analysis of distribution of metals in different size fractions did not indicate any patterns suggesting a different origin of these metals at different locations. References: Regulations about allowed quantities of d
Southeastern United States: Contrasts in Sources and Health Associations between
Inorganic Ions, Organic Ions, and Organophosphate Ester Oxidation Potentials

The use of oxidant species, such as hydrogen peroxide, to decompose PM samples is a widespread practice to determine whether they can affect human health. Inorganic ions, organic ions, and organophosphate esters (OPEs) have been identified as important PM components. OPEs have been suggested to play a role in the generation of reactive oxygen species (ROS). However, there is a lack of consensus on the role of these compounds, and their contribution is difficult to assess. The aim of this study was to determine the relative contribution of OPEs and inorganic species to the formation of ROS in PM samples collected in different locations in the Northeastern United States. The samples were analyzed using DTT, AA, and 2′,7′-dichlorofluorescin (DCFH).

Methods

Samples were collected in October 2016, from locations in the northeastern United States. The samples were extracted with 10% trichloroacetic acid and analyzed using DTT, AA, and DCFH. The results were compared to previously published data to determine the relative contribution of OPEs and inorganic ions to the formation of ROS.

Results

The results showed that OPEs were a significant contributor to the formation of ROS in the samples. The contribution of OPEs was highest in the samples collected from urban locations, while in the rural samples, inorganic ions were more significant. The contribution of OPEs was highest in the samples collected from urban locations, while in the rural samples, inorganic ions were more significant. The contribution of OPEs was highest in the samples collected from urban locations, while in the rural samples, inorganic ions were more significant.

Discussion

The results of this study highlight the importance of OPEs in the formation of ROS in PM samples. The significance of OPEs in the formation of ROS has been previously observed in other studies, but this study provides evidence for the contribution of OPEs in the formation of ROS in PM samples collected in different locations in the northeastern United States. The results also suggest that OPEs are a significant contributor to the formation of ROS in PM samples collected in different locations in the northeastern United States. The results also suggest that OPEs are a significant contributor to the formation of ROS in PM samples collected in different locations in the northeastern United States.
Challenges in setting, meeting and measuring specific protection goals for plant protection products (P)

**TU375**
French Phytopharmacovigilance: a national scheme for monitoring the adverse effects of plant protection products

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Phytopharmacovigilance is the latest complement to ANSES’s existing missions concerning the assessment of the risks associated with plant protection products before marketing, and the issuing and withdrawal of marketing authorizations. It is also fully in line with the third component of the Ecophyto plan. This national plan, established for the first time in 2008, was recently renewed. It aims to reduce pesticide uses in accordance with the requirements of Directive 2009/128, establishing a framework for Community action to achieve the sustainable use of pesticides. A specific scheme for funding these studies is planned through a tax on sales of plant protection products payable by the marketing authorization holders. To meet this objective, phytopharmacovigilance relies on three fundamental and complementary methods of data collection and knowledge production: a network of surveillance or vigilance bodies, collection of spontaneous reports and ad hoc studies on the adverse effects of plant protection products. These studies are financed by IPV to meet three different needs: 1) when the information provided by the surveillance and vigilance bodies is seen to warrant clarification, 2) to investigate spontaneous reports or 3) to collect new data / information. To identify the adverse effects of plant protection products on biodiversity and ecosystems, Phytopharmacovigilance is based on the systematic and regular collection of information produced by the existing surveillance and vigilance bodies, covering risks and impacts on wildlife, crops, fauna, flora, air, water, and soil. In the full article (2 pages word/pdf), a detailed list of studies and network working on “Pesticides impacts on biodiversity” and “Monitoring of pesticides (water, air, etc.,...)” is described.
schemes at EFSA and how recovery could be assessed considering the complexity of the environment. An important aim of these activities was to promote a dialogue between different panels of EFSA and risk assessors and risk managers responsible for the food and feed chains. Another important aim was to provide risk assessors with a conceptual framework to address ecological recovery in ERAs for assessed products, and invasive alien species that are harmful for plant health. This framework proposes an integrative approach based on well-defined specific procedures and guiding principles to allow the assessment of potential benefits and likelihood to develop environmental scenarios to allow the assessment of recovery of organisms and ecological processes at relevant spatial and temporal scales. Due to the complexity of ecological systems and the need to evaluate effects and risk, integrated systems in space and time caused by natural and human-induced anthropogenic factors, systems approach is required. The systems approach allows the integration of the various species, environmental factors, scales, and stressor-related responses necessary to address the context dependency in ecological recovery. The presentation will highlight the most important conclusions, challenges and recommendations to appropriately address ecological recovery in ERA for potential stressors that fall under the remit of EFSA EFSA Scientific Committee. 2016. Recovery in environmental risk assessments at EFSA. EFSA Journal 2:016; 14(2):4313. 85 pp

TU380 Habitat Equivalency Analysis for a Restorative Restoration Model of the Rio Doce Basin

P.N. Boech, Ramboll Environ / Ecological Sciences; E. Singer, F. Gomes, R. Arantes, Ramboll Environ / Sao Paulo; R. Wenning, Ramboll Environ

A Restorative Equivalency Model was prepared to support environmental, economic, and social restoration of the Rio Doce Basin after the Fundão Dam failure in Mariana, Minas Gerais State, Brazil. A Habitat Equivalency Analysis (HEA) was applied to quantify lost ecosystem services resulting from the disaster and was intended as a method to scale the potential habitat restoration of the Rio Doce to be refined through a process of stakeholder engagement to determine the valued environmental components (VECs); and thus the valued ecosystem services that should drive the restoration goals for each reach of the Rio Doce River. HEA is a well-accepted spatially and temporally explicit method for integrating multiple complex and difficult to measure environmental variables into a few metrics to determine overall losses and gains in ecosystem services resulting from impacts or restoration actions. The HEA method is applicable to any ecosystem and its flexibility allows for variability in the valuation of ecosystem services between communities and cultures. This paper focuses on the development and application of HEA within the context of the Resilience Model, and how selection of VECs as indicators to guide the scale, and location of restoration is aimed at developing an overall ecosystem restoration program that is at once cost-effective and results in a more resilient Rio Doce basin.

TU381 Using risk and recovery information in environmental cost-benefit analysis for determining appropriate risk management actions at major industrial facilities

A.E. Bartram, Ramboll Environ / Product Safety, Ecology and Sediment Management; S. Deacon, Ramboll Environ / Health Limited

Operators of chemical manufacturing plants and fuel storage depots are required to undertake site safety assessments, with specific emphasis on the control of major accidents and scaling the potential habitat restoration. HEA is being applied to identify an appropriate recovery duration based on potential recovery of key habitat types or species. This includes consideration of key factors affecting recovery and the fate and effects of SEVESO III chemicals, such as PBT substances with non-standardised spill containment systems. The sites are in close proximity to water bodies, the coast, and protected conservation areas. Site safety reporting requires an initial risk assessment be undertaken. The assessment draws upon both the potential severity of harm and environmental recovery in order to determine the risk tolerability under accidental release scenarios. Industry guidance exists on the evaluation of harm, but corresponding guidance on the prediction of environmental recovery was, until recently, limited. On behalf of the Energy Institute, Ramboll Environ developed a guide for risk assessors to determine the environmental recovery duration following major accidents of releases of SEVESO substances. Published October 2017, the guide provides a step-wise framework to identify an appropriate recovery duration based on one of four scenarios and the types of event involved. This includes consideration of key factors affecting recovery and the fate and effects of SEVESO III chemicals, such as PBT substances with the potential for longer-term impacts/delayed recovery. If an assessor determines that risks are intolerable, then the regulator requires facilities to consider appropriate investment in order that risks are as low as reasonably practicable.

Ramboll Environ has pioneered a methodology based on ecosystem services concepts to evaluate the costs and benefits of potential risk management options. The method is used to evaluate the risk in terms of potential damage avoided by putting risks into a socio-economic context. Case study examples will be provided that a range of infrastructure upgrade options at fuel storage depots are compared. Environmental Cost Benefit Analysis is used to determine if the upgrade would be grossly disproportionate to the benefit that would provide it. This analysis incorporates site-specific baseline ecology, other receptors and ecosystem services provided to society. The poster will also reflect upon lessons learned by both regulators and industry in the method development. References: Energy Institute (2017). Guide to predicting environmental recovery durations from major accidents. Supporting guide to the Environmental risk tolerability for COMAH establishments guideline

TU382 Addressing Resilience in Ecosystem Services Assessment

K. Mokhoviri, Ramboll E&H / Ecological Services; R. Wenning, Ramboll Environ / Ecology & Sediment Management; E. Bizzotto, H.R. Diogo, Ramboll / Ecological Services

An ecosystem services approach to landscape and nature restoration planning and damage assessment should fully account for all aspects of the environment and the human well-being derived from protection, enhancement and repair to natural resources caused by events and consequences of damaged events. With respect to damage assessment, conceiving of a resource as a part of an ecosystem that supplies valuable goods and services to people provides a basis for measuring changes and for valuation of those goods and services. With respect to landscape and nature restoration planning, an ecosystem services approach can lead to innovative ideas for ecological infrastructure in cities and promote ecological rehabilitation that may be ecologically and socially desirable and also economically advantageous. Resilience, however, is a key consideration that, to date, has been inadequately considered in ecosystem services assessment work. Considerations of resilience are especially important in ecosystems, because increasing resilience can reduce the risk that highly valued goods and services will cross critical thresholds and irreversibly degrade or change. Resilience also plays an important role in multiple complex and difficult to measure environmental variables into a few metrics to determine overall losses and gains in ecosystem services resulting from impacts or restoration actions. The HEA method is applicable to any ecosystem and its flexibility allows for variability in the valuation of ecosystem services between communities and cultures. This paper focuses on the development and application of HEA within the context of the Resilience Model, and how selection of VECs as indicators to guide the scale, and location of restoration is aimed at developing an overall ecosystem restoration program that is at once cost-effective and results in a more resilient Rio Doce basin.

TU383 Use of cost modelling techniques to manage environmental subsurface risks, liabilities and uncertainties in Spain

P. Wouters, M. Ferreira, I. Harper, Ramboll Environ / Environment and Health; . Salinas, Red ELÁctica de España

Companies owning large portfolios of properties are often faced with a high degree of uncertainty in relation to the subsurface conditions of their sites. This makes it difficult, if not almost impossible, to develop and implement a rational and cost-effective strategy to manage their regulatory obligations and financial liabilities. A customised probabilistic risk model was developed to facilitate the management of environmental and reputational risks at a portfolio of over 500 industrial sites in Spain. The sites are equipped with industrial equipment that contains large amount of oils. The equipment differs significantly in age, size, depth and make, and is equipped with non-standardised spill containment systems. The sites are distributed throughout the Spanish mainland and in the Baleares and Canary islands, and their environmental and social settings show a large variability. Site investigations were known to have been performed at only 5% of the sites. Initially an Environmental Risk Assessment Model was developed using technical, regulatory, social and environmental data. The compiled and consolidated data were entered into a calculation model developed in Microsoft Office Excel. Monte Carlo simulation was used to manage the potential range of scenarios that could be associated with particular assumptions in the model. Oracle®’s Crystal Ball® add-on to Excel was used to assign probability distributions to such uncertain model inputs. Probabilistic inputs were considered as risk scenario triggers for specific events at specific sites, such as: the likelihood of historic contamination being detected; a new contamination event being generated either on-site or off-site; a subsoil investigation being triggered; soil remediation being required; active or passive groundwater remediation being required; and implementation of a groundwater monitoring programme being required. Probabilistic inputs were also applied to the various cost scenarios that might be triggered. The model generated an environmental risk ranking expressed in purely financial terms. Ten high risk and 23 moderately-high risk sites were identified and an environmental action plan focusing on these highest priority sites was prepared. This allowed the portfolio owner to direct financial and human resources required for site investigation, remediation and preventive maintenance to those sites which could give rise to the highest financial and reputational liabilities.

TU384 Quality stakeholder involvement for resilience in environmental risk

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SETAC Europe 28th Annual Meeting Abstract Book
Assessment of Indoor Radon Concentration and Trace Metals Composition in University Building Microenvironments

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This study measures the indoor particulate matter (PM2.5) concentration and the equilibrium equivalent radon (EECN) concentration in two university buildings with different ventilation systems. A low volume sampler using Teflon filter paper was used to collect the PM2.5 samples and inductively coupled plasma mass spectrometry was used to determine the concentration of heavy metals. The concentration of indoor radon was measured using a radon detector model DOSEMann PRO. The potential human health damage due to the inhalation of carcinogenic and non-carcinogenic trace elements was also determined based on the US EPA standard. The equilibrium factor and the annual effective dose on the lung cancer risks of each occupant were calculated and a correlation of the radon concentration was made with the annual inhalation dose of the occupants at the outdoor stations. The results showed PM2.5 concentrations recorded in Building 1 and Building 2 ranged between 19.1 to 237 µg m⁻³ and 23.4 to 159 µg m⁻³, respectively. In Buildings 1 and 2, the annual inhalation dose of radon in Building 1 ranged between 19.1 to 237 µg m⁻³ and 23.4 to 159 µg m⁻³, respectively. The effective lifetime carcinogenic risks (ELCR) in Buildings 1 and 2 were 0.92E⁻⁶ and 1.65E⁻⁵, respectively. The hazard quotient (HQ) represents the non-carcinogenic risk, with values of 7.73 and 6.46 in Building 1 and Building 2, respectively. The average equilibrium equivalent radon measured in Buildings 1 and 2 was 2.33 ± 0.99 and 3.17 ± 1.74 Bqm⁻¹, respectively. The average annual inhalation doses recorded at Buildings 1 and 2 were 0.014 ± 0.005 mSv y⁻¹ and 0.020 ± 0.013 mSv y⁻¹, respectively. For trace metals, the ECLR and HQ values were found to exceed the permissible limits suggested by US EPA, whereas the values of equilibrium equivalent radon concentration were still below the standard recommended by ICRP.

TU388
Paradigm for PM2.5 Chemical and Biological Characterization: Paired Home and Personal PM2.5 Samples in Kheri, India

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The global public health impact from household fine particulate matter (PM2.5) is extremely large however, there is a limited understanding of health effects associated with specific PM2.5 chemical constituents as well as the underlying mechanisms of these adverse health effects. These research gaps can be addressed through use of a high-throughput screening platform to quickly gain biological response data. A subset of homes in Kheri, India that participated in the Prospective Urban and Rural Epidemiology (PURE) AIR pilot study were selected to identify differences in chemical and biological measurements of household PM2.5. In 6 households, personal air monitors collecting PM2.5 were worn by female participants and paired with stationary monitors, resulting in personal (n=6) and home (n=6) PM2.5 filters for each household. PM2.5 was removed from filters via sonication in methanol. Aliquots of individual filter samples were removed for oxidative potential assessment. Remaining PM2.5 samples of the same collection method were pooled (n=6/group) and the soluble fraction of PM2.5 from DMSO extraction was prepared for development toxicity testing performed in zebrafish (n=32/treatment) starting at 6 hours post fertilization (hpf). Aliquots of the pooled samples were used for chemical analysis (polycyclic aromatic hydrocarbons (PAHs, n=20), elements (n=20)) and oxidative potential assessment with methods identical to those used for filter extracts. Significant differences were observed in oxidative potential between personal and home PM2.5 for both individual and pooled samples. Significant mortality in zebrafish was observed starting at 24 hpf in personal PM2.5 samples and by 120 hpf in home PM2.5 compared to blank filter controls. Chemical analysis is underway to allow for correlations to be investigated between these biological responses and chemical constituents. This research is the first study to use paired home and personal PM2.5 samples with chemical, oxidative potential, and developmental toxicity data, identifying the differences in these measurements between household and personal PM2.5. Importantly, it outlines procedures for large-scale analysis of the PURE-AIR study which includes planned PM2.5 measurements in 4,000 homes and will ultimately allow for correlation of human health effects with chemical and biological data to identify improved health metrics for PM2.5 exposures.

TU389
Toxicity of Airborne Particulate Matter as a Factor to Choose the Most Conventional School

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One of the critical decisions parents have to face is choosing a good school for their kids. Proximity, high ratio of teachers to students, and access to convenient educational infrastructures are the driving factors determining school’s choice. However it is used to assume that environmental air inside schools is safe enough.
Among the different air pollutants found in schools, PM_{2.5} (airborne particulate matter smaller than 2.5 µm; also referred as "fine PM") is considered as the most injurious one. Since this pollutant is potentially very harmful, toxicity of PM_{2.5} on lung cells has been widely studied. However, most of the publications on this topic are focused on studying PM_{2.5} effects on human alveolar cells for short periods of time after applying doses far higher than environmental levels. To surpass this gap, we conducted a present study. On it, we collected two fractions of fine PM (PM_{2.5-10} and PM_{<2.5}) in indoor classrooms of some schools located under the influence of three different environments: urban, petrochemical, and chemical. Subsequently, we exposed human alveolar epithelial cells (A549) to IC_{50} doses obtained from a previous study. Then, apoptosis and release of 17 cytokines from the cells was studied after five different exposure times: 0, 6, 24, 48, and 72 hours. Preliminary results showed no differences in cytokines release among the two PM sizes or three sampling sites. However, differences arose when comparing the levels of seven cytokines (MCP-1, TNF-α, INF-γ, G-CSF, IL-6, IL-7, and IL-8) versus exposure times. These differences became significant after 24-48 h from exposure, and increased till reaching the maximum value after 72 hours. Results from this study will be useful not only to better understand the way of action of PM_{2.5}, but also to schools managers and parents.

TU390
Acute Impacts of Extreme Hot Temperature Exposure on Emergency Room Visits in Southern California
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Abstract The purpose of this study was to assess the effects of extremely high air temperatures on hospital emergency room visits (ER) related to alcohol addiction and other mental illnesses in Taiwan. A time series study was conducted using health and climatic data from 2000 to 2010 in Taiwan. A national health insurance database, temperature database, and air quality surveillance database were used for this study. Relative risks (RRs) for increases in emergency room (ER) visits were estimated for alcohol addiction and other mental illnesses after exposure to extremely high temperatures (99th percentile) and the 50th percentile of the daily mean temperature as reference. Poisson regression models using a distributed lag non-linear model (DLNM) were used. We adjusted for the effects of humidity and outdoor air pollutants. We found an association between alcohol addiction and other mental illnesses and mean daily temperature at 23.6°C on ER visits. The association was strongest within 0–7 days after exposure to higher temperatures. Increases (RR 1.02, 95% CI 1.01–1.04) in a major depressive disorder (MDD) ER visits was observed over a cumulative period of 7 days after exposure to high ambient temperature (99th percentile vs. 50th percentile). The opposite association was reported for alcohol addiction (RR 0.99, 95% CI 0.98–0.99). No significant associations with anxiety, dementia, and delirium were estimated. Our findings suggest that extreme temperatures pose a risk to the health and wellbeing for individuals with alcohol addiction and other mental illnesses.

TU391
Characteristics of Polybrominated Diphenyl Ethers Released from Primitive E-Waste Treatment
J. Li, Jinan University; J. Zhou, C. Wu, L. Bao, L. Shi, E.Y. Zeng, Jinan University / School of Environment
Abstract Primitive processing of e-waste potentially releases abundant organic contaminants to the environment, but the magnitudes and mechanisms remain to be adequately addressed. The present study conducted thermal treatment and open burning of typical e-wastes, i.e., plastics and printed circuit boards. Emission factors of the studied materials were assessed by exposing diphenyl ether and the upwind/ downwind ratios of PM_{10} (PM_{5-10}) and RL_{9.14} × 10^{7} g m^{-3} in thermal treatment and open burning, respectively. Airborne particles (87%) were the main carriers of PBDEs, followed by residual ashes (13%) and gaseous constituents (0.3%), in thermal treatment, while they were 30%, 43% and 27.2% in open burning. The input-output mass ratios of ∑_{i} PBDE were 0.21-10 in thermal treatment and 0.01–0.36 in open burning. All PBDEs were largely affiliated with fine particles, with geometric mean diameters at 0.61–0.83 µm in thermal degradation and 0.57–1.16 µm in open burning from plastic casings, compared to 0.44–0.56 and RL_{0.55} µm from printed circuit boards. The main emission mechanisms for lightly and heavily brominated BDEs were suggested to be evaporation and mechanical fragmentation, respectively. The difference between the size distributions of particulate PBDEs in emission sources and adjacent air implicated a noteworthy redispersion process during atmospheric dispersal.

TU392
How risky is the schoolyard? An approach from chemical composition of particulate matter
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Abstract According to last estimations, there are globally around 6.5 million deadliest a consequence of exposure to air pollutants. Among them, Particulate Matter (PM) is considered as the most harmful one. This material consists on solid particles and liquid droplets suspended in the atmosphere having a diameter smaller than 10 µm. Since they can come from a wide array of different sources (i.e. traffic, industries, indoor dust) their physicochemical characteristics are very heterogeneous, and knowing them becomes important in order to assess its damaging potential. To improve the knowledge regarding physicochemical characteristics of PM that children are exposed in schools we conducted a study focused on finding out chemical characterization of PM_{2.5}, PM_{10}, and PM_{1} (i.e. particles smaller than 10, 2.5 and 1 µm respectively) in an industrial area in Tarragona (Spain). These three fractions of PM were collected in the schoolyard (high volume samplers Thomas DV-7000, Tisch) and inside the classroom (low volume Sioutas cascade impactor, SKC) of 12 schools during two seasons (winter and summer). Subsequently, chemical characterization of the particles (through the analysis of metals, soluble ions and carbonaceous materials) was performed. Preliminary results show that both indoor and outdoor levels of PM are higher in winter than in summer. Indoor/outdoor ratios of particles are varying, showing some schools higher PM levels indoors, while the opposite phenomenon is observed in others. Consequently, concentrations of the different chemical constituents of particles are variable, although indoor particles trend to have higher shares of carbonaceous materials. Our results will be useful not only to schools managers and parents, but also to policy makers in order to assess risk coming from the inhalation of these materials.

TU393
Good news to lazybones kids: increasing sleeping time decreases exposure to airborne particulate matter
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Abstract The aim of the present study was to assess the children’s exposure to PM_{2.5} and PM_{10} and their influences on their activity patterns, the indoor and outdoor heat load, and outdoor air pollutants. The children’s airway exposure to PM_{2.5} and PM_{10} was evaluated in 12 schools of one of these 12 schools within the Tarragona county (Spain), an area characterized by having one of the most prominent industrial clusters in southern Europe. To elucidate time-activity patterns of kids, and to know the characteristics of their dwellings, questionnaires were delivered to parents of kids attending these schools. Using an infiltration model (IAQX, US EPA) it was possible to calculate concentrations of PM inside houses. A subsequent run of a dosimetry model (MPD2.11, ARA) made possible to know the deposition pattern of the different PM sizes within the distinct parts of the respiratory tract. Indoor/outdoor ratios of PM levels were variable among schools. Half of the schools presented higher concentrations of PM indoors, while the other half showed the opposite trend. Simulated the great influence of PM_{2.5} indoors, due to its easier capacity of infiltration from outdoors. Despite sleeping was the most time demanding activity, deposition fractions into the lung during these sleeping hours reached the minimum values. On the other hand, although moderate and high intensity activities accounted for 25% of time, these activities were responsible for the retention of 50–75% of overall PM mass. Most of this mass was deposited in the inspiratory tracts (98%), mainly due to sedimentation processes. Tracheobronchial region registered the lowest values of deposited particles, while PM retained in the lung was mostly PM_{10} and PM_{1}. Results from this study will help to take actions regarding indoor air quality and perform more accurate risk assessment studies.

TU394
Occupational Cement Dust Exposure: effect on blood level of some antioxidant enzymes and vitamins in Owerri, Nigeria.
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Abstract Despite that Cement dust with particulate matters are hazardous to humans, yet Occupational cement dust exposure continues among Cement workers. The workers in cement industries are exposed to high levels of some antioxidant enzymes and vitamins have not been adequately addressed especially in a black-African environment and particularly Nigeria. By random sampling method, 35 Cement workers, 35 Cement Dealers and 35 controls were selected for this study. Blood antioxidant enzyme such as glutathione peroxidase, superoxide dismutase (SOD) and catalase (CAT) were determined using ELISA while antioxidant vitamins such as vitamin E and vitamin C were determined by spectrophotometric techniques. There were progressive significant increases in blood level of vitamin C, vitamin E, catalase and glutathione peroxidase from Cement Workers to Dealers and Controls (non-cement workers), (P>0.0010, P>0.0011,
P=0.0011) respectively. There were significant decreases in blood levels of vitamin C, vitamin E, glutathione peroxidase and catalase (P<0.002, P=0.0004, P=0.0011) respectively in Cement Workers/Dealers compared to Controls. But no significant difference in SOD (P=0.627) on comparison of Cement Workers/Dealers with Non-Cement Workers (Controls). There was significant decrease in blood levels of vitamin C (P=0.0147), SOD (P=0.0256), glutathione peroxidase (P=0.015) and vitamin E (P=0.058, r=0.256 and r=0.13) respectively, but there was a non-significant positive correlation of vitamin E with; Catalase, SOD, Glutathione peroxidase and Vitamin C, (r=0.256, r=0.315, r=0.04598 and r=0.208 respectively). There were non-significant correlations of catalase with glutathione peroxidase, vitamin E, and vitamin C (r=0.058, r=0.256 and r=0.13 respectively), but there was a positive significant correlation of catalase with SOD (r=0.4173). This study suggest that Exposure to cement Dust may lead to reduction in blood levels of vitamin C, Vitamin E, Catalase , Glutathione Peroxidase, and SOD in Cement workers/Dealers. The observed reduction/decreases in the antioxidants were progressive from control to Cement Dealers and to cement Workers. Key words; Cement dust, antioxidant, enzymes, vitamins.

TU395 Implementing NH3 mitigation strategies in a pig farm: different approaches to evaluate the environmental impact. C. Baldi, Università degli Studi di Milano / Department of Environmental Science and Policy; J. Bacenetti, Università degli Studi di Milano / Department of Environmental Science and Policy; F. Fermo, Università degli Studi di Milano / Department of Chemistry; M. Guarino, Università degli Studi di Milano / Department of Environmental Science and Policy.

Ammonia is an atmospheric pollutant causing acidification of soil, nutrient-N enrichment of ecosystems, and eutrophication of terrestrial and aquatic ecosystems. When in gaseous form, NH3 has a short atmospheric lifetime and usually deposits near its source. In the atmosphere it reacts with other compounds to form ammonium sulfate and ammonium nitrate aerosols, leading to the formation of secondary inorganic aerosol (PM2.5) that are a potential health hazard. Due to their smaller diameter and increased atmospheric lifetime, these particulates are able to travel long distances before being dry or wet deposited to the ground surface. This allows them to travel from rural areas to urban locations where they mix and build up in the atmosphere leading to smog or transportation to other areas. The particular unfavorable meteorological and orographic conditions of the Po Valley make this one of the most polluted region of Europe. Particulate matter pollution often exceeds the EU standards and WHO air quality guidelines for health protection. But a main source of ammonia emissions, the agro-zootechnical compartment plays a key role in the secondary PM formation. Indeed, secondary inorganic aerosol from NH3 accounts for 40% of PM2.5 mass at the urban sites, and its contribution is even bigger in the rural sites. This study aims at evaluating the environmental performance of different NH3 mitigation strategies applied to Italian pig farms. Different mitigation scenarios are compared, considering the application of odor control strategies in the manure and to the manure ammonification and the application of odor control strategies in the manure and to the manure storage. The studied toxic potentials were elicited mainly by chemicals in the polar fraction containing relatively high levels of oxygenated-polycyclic aromatic hydrocarbons (oxy-PAHs). This study confirms that several molecular mechanisms of toxicity are associated with air pollution and highlights the complexity of pollutant mixtures. For further understanding, the results will be discussed together with the results of the chemical analysis which focused on PAHs and their derivatives, nitro- and oxo-PAHs. This research was supported by project GACR P503 16-11537S.

TU396 Development of an In Vitro Method to Evaluate the Inhalation Bioaccessibility of Particle-Bound Hydrophobic Organic Chemicals and its Effects of Particle Size. S. Liu, Jinan University; L. Bao, E.Y. Zeng, Jinan University / School of Environment.

Bioaccessibility of particle-bound hydrophobic organic contaminants and its effects of particle size are significant for assessing the potential human health risk via inhalation exposure, but have not been clearly evaluated. To fill this knowledge gap, the present study develops an in vitro method to estimate the inhalation bioaccessibility of particle-bound hydrophobic organic contaminants using lung fluids, i.e., artificial lysosomal fluid (ALF) and Gamble’s solution amended by dipalmitoyl-sn-glycerol-3-phosphocholine, with Tenax as the absorption sink. Polycyclic aromatic hydrocarbons (PAHs) were selected as the target compounds and the assay parameters such as incubation time and the influence of particulate load membrane were examined. Results have shown that the bioaccessibilities of individual PAH compounds increased with the increasing incubation time and reached the steady state within 10 days. None of significant difference was found for the individual PAH bioaccessibilities between with and without adding glass microfiber membrane into the incubation system. Furthermore, the PAHs absolute recoveries, calculated by sum of PAHs masses in Tenax, artificial lung fluid, and residual particle dividing the initial masses, were from 92% to 112% in ALF and 75% to 99% in Gamble’s solution, suggesting that this developed in vitro method could be well appropriate to evaluate the inhalation bioaccessibility to particulate hydrophobic organics matter. In addition, the PAHs bioaccessibility were found to be increased with particle size, but decrease with the increasing hydrophobicity. It is necessary to separate the influence of the inhalation exposure from the size effect. The size-dependent PAHs bioaccessibility was reduced by more than 90% if the size-dependent PAHs bioaccessibility and deposition efficiency were involved into the assessment.

TU397 Toxicity does not vanish into thin air - molecular mechanisms of air pollutant mixtures. Z. Novakova, Masaryk University; J. Novak, Masaryk University / RECETOX; K. Hilscherova, Masaryk University, Faculty of Science, RECETOX / Research Centre for Toxic Compounds in the Environment RECETOX; P. Kakucka, P. Pribylova, P. Prokes, Masaryk University / RECETOX; G. Lammel, Max Planck Institute for Chemistry / Multiphase Chemistry Department.

Air pollutants remain to have adverse effects on human health and can directly affect human health and can contribute to higher incidence of pulmonary and cardiovascular diseases or cancer. Moreover, it has been shown that compounds occurring in the ambient air pose a hazard to disrupt our endocrine, immune or reproductive system. Since the air pollutants occur in complex mixtures, it is more appropriate to use an effect-based monitoring including a battery of in vitro bioassays which cover various interactions among mixture constituents. Bioassays represent an efficient approach for toxicological profiling and identification of pollutant modes of action. Together with chemicals analyses, they enable to identify main toxicity drivers. Two sites were selected, a heavily polluted urban site (industries, transportation, coal-based domestic heating) and a regional background. Samples were collected in summer and winter. To assess the more specific contribution of the fine PM (PM2.5) to the health impact, coarse particulate phase, and six PM2.5 size fractions were sampled. Moreover, samples were also fractionated according to polarity. Human-based in vitro bioassays were employed to study endocrine-disruptive potentials, AhR-mediated induction of detoxification mechanisms, and cyto- and genotoxicity to the human respiratory tract. The results show that the studied effects were associated mainly with particulate phase. The most significant effects were attributed to the easily inhalable fine and ultrafine particles. This distribution pattern was found for example for AhR-mediated toxicity, estrogenicity, and androgenicity. The studied toxic potentials were elicited mainly by chemicals in the polar fraction containing relatively high levels of oxygenated-polycyclic aromatic hydrocarbons (oxy-PAHs). This study confirms that several molecular mechanisms of toxicity are associated with air pollution and highlights the complexity of pollutant mixtures. For further understanding, the results will be discussed together with the results of the chemical analysis which focused on PAHs and their derivatives, nitro- and oxo-PAHs. This research was supported by project GACR P503 16-11537S.

TU398 Human health assessment of air pollution exposure to tuberculosis risk in regions of Taiwan. H. Lin, National Taiwan University; Y. Lin, National Health Research Institutes / National Institute of Environmental Health Sciences; Y. Yang, C. CHEN, National Taiwan University / Bioenvironmental Systems Engineering; C. Liao, National Taiwan University / Department of Bioenvironmental Systems Engineering.

BACKGROUND: Ambient air pollutants from anthropogenic sources play critical roles in the prevalence of tuberculosis (TB) risk. In regions of Taiwan. Additionally, the particulate matter (PM2.5) and nitrogen oxides (NOx) also were likely to contribute to TB incidences in some regions. CONCLUSIONS: We suggested that the contributions of air pollutants mainly from diesel combustions (CO, NOx and NOy) to TB incidences are of great concern. Furthermore, the human health risk assessment framework provides an alternative perspective to interpret the effects of air pollution on TB burdens. Keywords: Human health risk assessment; Air pollution; Tuberculosis; Population attributable fraction; Probabilistic risk assessment.
Towards green braking: comparative evaluation of toxicological profile of particles generated by traditional and innovative braking systems.

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The protection and improvement of air quality are key critical points of environmental protection, health, and international level. Inhalation of contaminated air and airborne particles is definitely the main route of exposure to the most important pollutants, secondary only to the diet. Several respiratory and cardiovascular diseases are associated with air pollution. Air pollution is responsible for 400,000 deaths per year in EU28. In 2013 outdoor air pollution was classified as carcinogenic to man (Group 1) by the International Agency for Research on Cancer, with a special attention to airborne dusts. In addition, powders can alter aquatic and terrestrial ecosystems when they reach the surface waters and soils as a result of transport and deposition events. Road transport and traffic contribute greatly to emissions of PM2.5 and PM10 and an important contribution to the non-exhaustive emission is due to the wear of brakes. The European Life+ COBRA (LIFE13 ENV/IT/000492) project aims to create a safer alternative to the previous brake design. The project’s goal is to replace the plastic brake material with a new cementitious hydraulic binder. The study here presented evaluated the eco- and toxicological potential of particulate matters generated in laboratory conditions using test benches capable of simulating vehicle braking cycles. PM2.5, PM10 and deposited non-airborne particles were collected. Fine and coarse particles were detached from the filters and dispersed in an alcohol solution for in vitro assessment of their toxicological potential with non-tumour bronchial epithelium BEAS-2B cells as model of the respiratory tract. Effects on cell proliferation and cytotoxicity were assessed daily at the MTS assay and the evaluation of DNA-release from damaged cells. Sublethal responses were also measured including oxidative stress, DNA damage, mitochondrial membrane potential and metabolic alterations.

In order to evaluate the ecotoxicological effects on seedlings growth and root damages, cress seeds were exposed to standard soil contaminated with non-airborne particles. The innovative cementitious pads were comparable to traditional pads in terms of braking performance but showed significantly reduced airborne particles production. Results showed different toxicity between particles generated from traditional and innovative braking systems: in our experimental models, particles derived from cementitious pads were less toxic than those from the resin based pads.

TU400 Toxic oxidation transformation products of phenanthrene measured in laboratory generated secondary organic aerosol particles
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Oxidized transformation products (OTP) of polycyclic aromatic hydrocarbons (PAHs) found in soils, suspended aerosols, and in small industrial areas located in the area. To this purpose, the approach based on the use of PM samplers coupled with a wind-select sensor, allowing a selective PM10 and PM2.5 sampling downwind to specific emission sources, has been used. Furthermore, the chemical characterization of the PM collected has been carried out in order to explain specific emission patterns, and to assess the concentration levels of the micro-pollutants emitted by local sources and particularly toxic for health. Two sampling sites have been identified in the area, respectively urban and urban background site, and equipped with a PM10 and PM2.5 wind-select sampling device, designed to collect aerosols from two directions, downwind respectively the coal-fired power plant and the port area. Samples of PM10 and PM2.5 were monthly collected for one year, and chemical analyses were performed to determine the concentrations of organic and inorganic species.

A descriptive-statistical analysis of data was performed, also verifying the occurrence of legislative threshold exceedances. Moreover, in order to highlight the contribution of specific sources, the differences in the measured micro pollutants concentrations between wind directions, PM size fractions and sampling sites have been investigated, as well as the seasonal trends of pollutants concentrations. These results allowed to highlight that the applied methodology represents a valid support in source apportionment studies. Keywords: source apportionment, wind select-sampling device, PM10, PM2.5

TU403 Forecasting global atmospheric visibility based on air quality and meteorological data
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Simultaneous and continuous measurements of visibility, meteorological parameters, including relative humidity, and the concentrations of six atmospheric pollutants (PM2.5, PM10, SO2, NO2, CO and O3) were obtained for several cites around the globe. The characteristics of visibility and relationships with air pollutants and meteorological factors were investigated using multiple statistical methods. Analysis demonstrated that within a certain relative humidity range, visibility is the exponential function of the PM10 concentration. Thus, non-linear models combining multiple linear regressions with exponential regression were subsequently developed to describe the hygroscopic growth and the attenuation effects on the air pollution visibility. The validation of these models demonstrated that the derived models can quantitatively describe the relationships between visibility, air quality and meteorological parameters around the whole globe.

TU404 Analyzing the Asian supply chain structure of health impacts with PM2.5 including secondary particulate
F. Nagashima, Kyushu University; K. Nansai, National Institute for Environmental Studies; S. Chatani, National Institute of Environmental Studies; S. Kagawa, Kyushu University

Analyzing the Asian supply chain structure of health impacts with PM2.5 including secondary particulate
Countries and regions in Asia have played an important role in producing intermediate products and final commodities today and supplied their products around the world. Productions and consumptions of goods and services in the Asian countries have clearly contributed to the economic growth in the world, whereas those economic activities brought about environmental loads. Particularly, China’s emissions of air pollutants such as fine particulate matter (PM<sub>2.5</sub>) have caused several serious environmental and public health problems in China; current research is focusing on identifying the health impacts associated with the PM<sub>2.5</sub> through the Asian supply chains have been estimated in the previous researches. While these analyses showed what production activities induced these health hazards caused by “primary” PM<sub>2.5</sub>, almost of these results doesn’t include the effects of “secondary” PM<sub>2.5</sub>. This study developed the secondary PM<sub>2.5</sub> concentrations emitted on every industrial sector based on the Emission Sources Database for Global Atmospheric Research (EDGAR) emission inventory data, Weather Research and Forecasting (WRF) model and CMAQ modeling system, and then estimates the induced mortality data in Asia. We further applied structural path analysis (SPA) to the Asian International Input-Output Table (AIIOIT) to identify the critical supply chains for the reduction of health impacts in Asia. The result shows that the Japan’s consumption contributed to secondary PM<sub>2.5</sub> emissions in Asian are estimated 6.35 Tg-C and we revealed top ranking supply-chain paths for PM<sub>2.5</sub> emissions induced by Japan’s final demand. The most significant supply-chain path with the highest emissions was the path from transportation sector in China to Japan’s final demand, and subsequently the path, other food products sector in Thailand --> food crops sector in Thailand --> Japan’s final demand. We also argued the health impacts caused by the trans-boundary pollutions in Asian countries.

TU405 Source contributions to PM10 levels in a coastal area in northern France: a one year study
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The Hauts-de-France Region is one of the most concerned areas in France by exceedances of the PM<sub>10</sub> daily mean limit value (50 µg/m<sup>3</sup>). For a better understanding of these phenomena, the identification as exhaustive as possible of source contributions to secondary PM<sub>10</sub> emissions in Northern France is estimated 1.05 Tg-C and the atmospheric water soluble ions, EC/OC as well as tracers of biomass burning (levoglucosan), collected using the DA80 sampler (Digitel®, 3 years in 2013 at Cape Gris Nez, a coastal French site located in front of the Straits of Dover). PM<sub>10</sub> sampling and measurement campaign were performed continuously during one year in 2013 at Cape Gris-Nez, a coastal French site located in front of the Straits of Dover. PM<sub>10</sub> levels were measured using MPI101 analyzer (Environment SA®) and collected using the DA80 sampler (Digitel®, 30 m<sup>2</sup>/h) on a daily basis. The characterization of PM<sub>10</sub> was performed considering major and trace elements, water-soluble ions EC/OC as well as tracers of biomass burning (levoglucosan), primary biogenic emissions (arabitol, mannitol) and marine biogenic emissions (methanesulfonate ions). These chemical parameters were used to explain PM<sub>10</sub> levels on the coastal site, identify PM<sub>10</sub> sources and estimate their contributions. Sources profiles were identified from the use of a Constrained Weighted non Negative Matrix Factorization (CNMF) model: fresh sea-salts, aged sea-salts, secondary nitrates, secondary sulphates, crustal, biomass combustion, primary biogenic emission, marine traffic, combustion, metal source. The monthly evolution of their contribution evidenced different behaviours between the sources: secondary nitrates were predominant during the cold season and appeared to be the most involved in the PM<sub>10</sub> concentration peaks. The impact of marine traffic and a high proportion of sea-salts versus fresh sea-salts was mainly evidenced during the summer season. For the year 2013, the mean contribution of the different sources were 37% for sea-salts and aged sea-salts, 43% for the secondary inorganic aerosols, 7% for biomass combustion, 5% for marine traffic. This distribution varies highly depending on the period and more particularly during exceedances of daily PM<sub>10</sub> limits values.

TU406 Source-to-exposure assessment of industrial pollutants in Australia, using the Pangea multi-scale framework
C. Wannaz, The University of Michigan, Ann Arbor / SPHEHS; P. Fantke, Technical University of Denmark / Quantitative Sustainability Assessment Division; J. Lane, University of Queensland, Brisbane; O. Jollivet, University of Michigan
Effective planning of airshed pollution mitigation is often constrained by a lack of integrative analysis able to relate the relevant emitters to the receptor populations at risk. Both emitter and receptor perspectives are therefore needed to consistently inform emission and exposure reduction measures. This presentation aims to extend the Pangea spatial multi-scale multimedia framework to evaluate source-to-receptor relationships of industrial sources of organic pollutants in Australia. Pangea solves a large compartmental system in parallel by block to determine arrays of masses at steady-state for 100,000+ compartments and 4,000+ substances. The framework uses a multi-mass approach and computes population exposure by inhalation and ingestion. From an emitter perspective, the spatial distribution of population exposures show high spatial variations in intake fractions from 0.68 to 33 ppm for benzene, and from 0.006 to 9.5 ppm for formaldehyde, contrasting urban, rural, desert, and sea emission source locations. Extending analyses to the receptor perspective, population exposures from the combined emissions of 4,101 sources in Australia were estimated for benzene, formaldehyde and see emission source locations. Extending analyses to the receptor perspective, population exposures from the combined emissions of 4,101 sources in Australia were estimated for benzene, formaldehyde and sea emission source locations. Extending analyses to the receptor perspective, population exposures from the combined emissions of 4,101 sources in Australia were estimated for benzene, formaldehyde and sea emission source locations. Extending analyses to the receptor perspective, population exposures from the combined emissions of 4,101 sources in Australia were estimated for benzene, formaldehyde and sea emission source locations.
Microplastic Indoor Air Pollution Using a Simulated Breathing Mannequin - µFT-IR Imaginary Quantification

N. van Alst, Aalborg University / Civil Engineering Department; A. Vianello, Aalborg University / Civil Engineering Department - Section of Water and Environment; R.L. Jensen, Aalborg University; J. Vollertsen, Aalborg University / Civil Engineering Department

Extensive research has been performed on indoor air quality (IAQ) over the last decades. This investigation focuses on the mechanisms of particle formation in indoor air and on surfaces, as well as the formation of microplastics. The mannequin takes air in through the mouth, which is led through a copper pipe to the filtering unit. The copper pipe meets a filter holder on which a 0.6 µm double-bowl metal shielding, there is no standardized design applied in all studies in terms of bowlder size, shape, deployment configuration. Many different PUF-PAS designs are used in regional or global programmes such as the Global Monitoring Programme under the Stockholm Convention and these data are compared for spatial and temporal trend analysis. Yet, no information is available on the comparability of data from all the different designs. We brought together 12 types of PUF-PAS samplers from around the world and deployed them in a multi-part cross-comparison in order to evaluate the variability in reported concentrations introduced by different elements of PAS monitoring. Three sets of PUF-PAS were deployed in Kjellera, Norway in 2015-2016, as follows: (1) 3-month deployment of 15 PAS provided by international research groups, and returned to their respective research groups for analysis for SVOCs – this provides information on the overall variability in global monitoring data introduced by differences in sampler configurations and analytical methods; (2) 3-month deployment of 15 identical PAS, which were then distributed to international laboratories for SVOC analysis, to isolate the influence of analytical variability; and (3) 3-month deployment of 15 different PAS and analysis at a single laboratory (RECTOX Trace Analytical Laboratories, Masaryk University) to isolate the influence of PAS design on data comparability. Results indicate that while differences in sampler design (in particular the spacing between the upper and lower sampler bowls) account for 50–100% differences in masses collected by samplers, the variability introduced by analytical methods still significantly exceeds this amount, and this effect should be carefully considered when evaluating and comparing global monitoring data.

Determination of Cross Compartment Concentration Gradients of Polycyclic Aromatic Hydrocarbons using PE Passive Samplers

N. van Alst, Aalborg University / Civil Engineering Department; A. Vianello, Aalborg University / Civil Engineering Department - Section of Water and Environment; R.L. Jensen, Aalborg University; J. Vollertsen, Aalborg University / Civil Engineering Department

Several polycyclic aromatic hydrocarbons (PAHs) are considered as human carcinogens or toxic to reproduction, and are thus a relevant class of “substances of very high concern” according to the European Chemicals Legislation REACH. Engine Place (PAHs) are naturally found in the environment, Polychlorinated biphenyls (PCBs) are considered a threat to human health and wildlife, and are listed as a substance of high concern in the REACH list. This is due to the potential for long-term exposure through the food chain, and their persistence in the environment. The aim of the present project is to measure the concentration of selected PAHs in the indoor environment of a high-rise building in a European city. The study was conducted in a high-rise building in a European city, where indoor air samples were collected using passive air samplers (PAS). The PAS devices were deployed in various rooms of the building, including kitchens, bathrooms, and bedrooms. The PAH concentrations were measured using a high-resolution gas chromatography/mass spectrometry (HRGC/MS) technique. The results showed that the indoor air concentrations of PAHs were significantly higher than the outdoor air concentrations. This indicates that indoor air may serve as a source of PAHs for the surrounding environment. The study also revealed that certain indoor activities, such as cooking and smoking, significantly increase the PAH concentrations in indoor air. Overall, the findings of this study highlight the importance of measuring indoor air PAH concentrations and identifying the sources of exposure in order to improve indoor air quality and health outcomes.
Kekulé resonance structures, 2) thermodynamic stability of all possible OH-PAH adduct intermediate, 3) electron density at each carbon on the PAH and 4) average local ionization energy (ALIE) at atom or bond sites. To evaluate the accuracy of these approaches, the predicted PAH-TPs were compared to published laboratory observations of major NPAH, OPAH, and OHPAH products in both gas- and particle-phases. We found that the Clar’s resonance structures were able to best predict the stable rings over the PAHs, but did not offer insights in terms of which carbon is most reactive. All other computational approaches provided specificity in their predictions, yet the ALIE approach was the most superior in accuracy, when compared to laboratory data. The high predictive capability of ALIE shows great potential for the prediction of the formation of previously unstudied PAH-TPs that are likely to form in the atmosphere. Furthermore, the results compared to the environmental chemists to prioritize which PAH-TPs might be formed in the environment; the organic chemists to prioritize which PAH-TPs should be synthesized to verify their presence in the environment; and the toxicologists to prioritize which PAH-TPs should be analyzed for their toxicity and potential human health implications. Future direction of the study is to expand the prediction to screen for PAH-TPs from other parent-PAHs as well as alkylated-PAHs.

TU413 Spatial distribution of gas-phase Polycyclic Aromatic Hydrocarbons along South America and Antarctica

A. Azrevedo, Instituto Federal do Rio Grande do Sul; K.S. Miglioranza, University of Minnesota, USA; J. Lao, FURG / Instituto de Oceanografia; E.M. Lopes, Instituto de Mar Del Plata / Laboratorio de Ecotoxicología y Contaminación Ambiental, Argentina; P.G. Costa, FURG / Escuela de química e alimentos; R. Barra, Universidade de Concepción; O.P. Amaranth Jr, IFMA / DAQ; F. Wania, University of Toronto at Scarborough / Physical and Environmental Sciences; G. Fillmann, Universidade Federal do Rio Grande / Instituto do Meio Ambiente; V. Azevedo, Universidade Federal do Rio Grande do Sul. Passive samplers and XAD2 resins were passed through the atmosphere employing XAD2-based passive atmospheric sampling (PAS). The Latin American Atmospheric Sampling Network (LAPAN) has begun in 2010 by deploying a pair of PAS containing one cartridge of XAD-2 resin on each site. Resins were deployed for 12 months during 3 consecutive years at42 sites (16 sites in Brazil and 26 distributed in Argentina, Chile, Peru, Uruguay, Venezuela and Antarctica) covering different backgrounds (rural, urban / industrial and remote). Passive samplers and XAD2 resins were prepared as described by Wania et al. (2003). XAD-2 resins were extracted by hexane:dichloromethane (1:1), purified and analyzed by gas chromatography/mass spectrometry (GC/MS). The following PAHs were analyzed: naphthalene, 2-methyl-naphthalene, 1-methyl-naphthalene, acenaphthylene, acenaphtene, fluorene, phenanthrene, anthracene, fluoranthene, pyrene, benzo(a)anthracene, chrysene, benzo(b)fluoranthene, benzo(k)fluoranthene, benzo(a)pyrene, indeno(1,2,3-cd)pyrene, dibenzo(ah)anthracene and benzo(g,h,i)perylene. Results, reported a distribution of gas phase PAHs along South America atmosphere.

TU414 Importance of Dermal Exposure to Polycyclic Aromatic Hydrocarbons Derived from Barbecue Fume

J. Lai, S. Xie, Jinan University, C. Wu, L. Bao, Jinan University / School of Environment; S. Tai, Tsing University / Laboratory for Earth Surface Processes College of Urban and Environmental Sciences; E. Y. Zeng, Jinan University / School of Environment

Despite the ubiquity and carcinogenicity of polycyclic aromatic hydrocarbons (PAHs), its dermal absorption for the general population has not been adequately addressed. Aiming to verify the importance of dermal absorption for PAHs, barbecued (BBQ) in China was chosen as the source. Unexposed samples were collected approximately 17 h before exposure until 35 h after exposure from 20 participants and analyzed for nine hydroxyl (OH)-PAHs. Air, food, and cotton clothing samples were analyzed for 16 PAHs. Based on the occurrence of atmospheric PAHs, dermal absorption of low molecular-weight PAHs was greater than inhalation intake. In addition, the net excreted amounts of OH-naphthalene, OH-fluorene, OH-phenanthrene, and OH-pyrene via dermal contact were 367, 63, 98, and 28 ng respectively, comparable to those via combined dermal and inhalation exposure, which were 453, 98, 126, and 38 ng. The ratios of excretion to intake via dermal contact were 0.11, 0.036, and 0.043 for fluorene, phenanthrene, and pyrene, respectively, higher than those for inhalation (0.097, 0.016, and 0.025). These results indicate that dermal absorption is a significant exposure route of PAHs. In the case of BBQ fumes, dermal absorption is a more important pathway for intake of low molecular-weight PAHs than inhalation.

TU415 EDS Mapping of Particles As A Component of Lichen Biomonitoring in Seattle, Washington

G.T. Guddal, Western Washington University; J. Miller, A. Johnson, Western Washington University / Environmental Sciences Department; R. Sofield, Western Washington University

Lichen are an increasingly popular medium for conducting air quality monitoring due to their sensitivity to SOx and NOx, as well as their biomonitoring of airborne material. This study incorporates characterization of particulate matter (PM) on the surface of lichen Ramalina farinacea to map exposure to air pollution in three industrial clusters in Seattle, Washington, USA. The PM was characterized using scanning electron microscopy with energy-dispersive X-ray spectroscopy mapping to determine PM size and composition. We also measured biomonitoring of certain PAHs in the lichen and the biomarkers of glutathione, chlorophyll degradation, malondialdehyde, and uric acid. Principal components analysis has identified which geographic locations and particle types correlate the strongest with increased metal accumulation and physiological response in the lichen.

TU416 TBARS in horse hair as an indicator of oil industry pollution

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Air pollution is a major problem today. Although there are many tests that measure the presence of certain substances in the air, it is important to measure the impact of various pollutants on living organisms as well. Horses that spend time outdoors are exposed to environmental influences, and some of them are measurable in horse hair. The aim of this study is to assess how pollutants of the oil industry affect biological markers in permanent horse hair from mane and tail and whether the concentration of thiobarbituric acid reactive substances (TBARS) can be used as a biomarker of air pollution. The horse hair samples from mane and tail were collected from two areas. One near Slavonski Brod, where an issue of air pollution is present due the outdated refinery plant in Bosanski Brod, and the other near Osijek where no apparent air industry pollution is present. The concentration of TBARS in samples was measured fluorometrically. The samples were cut into segments to detect differences in duration of exposure through the age of hair. The concentration of TBARS was significantly higher (p<0.005) in horses exposed to polluted air (Slavonski Brod site). When segments were analysed according to the age of hair it was noticed a constant difference in concentration of TBARS between roots and top for mane and tail. A significant correlation between age of hair and concentration of TBARS was noticed. Although further research is needed, a concentration of TBARS in horse hair could serve as a simple and inexpensive monitoring air pollution by oil industry.

TU417 Morbidity for environment-related diseases in La Spezia, northwest Italy: an epidemiological analysis on hospital discharge rates

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Background. La Spezia Province, northwest Italy, is a potentially high risk area because of the massive presence of industrial and harbor activities. Only controversial evidence of their health impact in the area is available at the moment. Therefore, a comprehensive survey on potentially environment-related disease would be really useful. Methods. Liguria Region hospital discharge records from 2001 to 2013 have been collected. Only admissions for cardiovascular disease, respiratory disease or neoplasms as primary diagnosis were included. Hospital discharge rates, standardized by age and compared with Regional mean, were represented using geographic maps with a color scale identifying different disease distribution. For those disorders showing significant difference with regional mean, disease distribution was compared with emissions of air pollutants, estimated by Lichens biomonitoring. Results. La Spezia Province Hospital Discharge rates for potentially environment-related disease were significantly different if compared with those of the Liguria Region. Malignant melanoma, chronic bronchitis and myocardial infarction rates were higher both in men and women (with a percentage increase of 84% and 87%, respectively, for malignant melanoma; +22% and +47% for chronic bronchitis and +40% and +41% for myocardial infarction). Conversely, hospital discharge rates for diabetes and hypertensive cardiopathy were lower than the Liguria Region: -50% among males and -49% among women for the first and -56% and -54%, respectively, for the second. Comparing these results with Lichens maps, no evidence of a clear correlation between emissions of air pollutants and regional distribution of diseases was available. On the contrary, diseases differing the most...
from Regional mean were mainly distributed in areas with scarce anthropic activity. 

Conclusions. No clear morbidity trend is identifiable for La Spezia Province from 2001 to 2013. Also disorders belonging to the same pathological class – like myocardial infarction and hypertensive cardiopathy - showed a different behavior. Comparing results with Lichen maps helps putting excesses of morbidity into a Regional context, being cautious to clearly correlate such disease with industrial and Harbour activity. Indeed, most of the disorders showed the higher increases in areas with less signs of anthropic activity, according to Lichens biomonitoring.

TU418 Risk Assessment of Polyethylene Residues and Organoleptic Attributes of Bambara nut pudding (Okpa) Samples prepared using Alternative Cooking Materials

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Polyethylene resins are chemical components that are left over as monomers and end products after the thermal degradation of polyethylene. However, the use of plastic as cooking materials in bambara nut pudding (Okpa) a well-cherished food, especially among the inhabitants of the Eastern part of Nigeria may pose more danger than envisaged. The use of banana leaves in cooking food has been in existence in Nigeria before the introduction of the technological cooking materials such as polyethylene, cellophane plastic, tin and foil. In this study, we evaluated the risk assessment of polyethylene residues (volatile organic compounds - VOCs) and organoleptic attributes of Bambara nut pudding (Okpa) samples prepared using alternative cooking materials. Purge and trap method using Gas Chromatography and Mass Spectrometry instrument were used to estimate the concentrations of VOCs in the different pudding cooked using some alternative cooking material (cellophane, tin, foil and plastic) while banana leaves were used as control. Organoleptic evaluation was done using A-Point Hedonic Scale, standard methods and ANOVA was used to compare means of the results. The result showed the presence of some Volatile Organic Compounds such as Argon, Allene, Acetic acid, Propene-1-ol, difluoromine, Hexanoic acid, Amyl nitrite, Toluene, Butenenitriile, 2-Butenal, Thiriane, Nonanoic acid, Ethylenediamine, Furfural, Hydrogen azide, 2-pentene, Formic acid, and octanoic acid; with Acetic acid occurring the most and Argon, Allene, and Difluoromine occurring the least. Pudding made with cellophane had the highest VOCs with 45% D-mannohptelolute, 45% hexanoic acid, 25% propane-1- ethylthilol and had other VOCs ranged from 4-9%. All the cooking materials had hexanoic acid at high concentrations of 25-42%. The result also showed that acetic acid and 2-butenenitrile ranged from 4-7% in all samples except Banana leaves pudding. Organoleptic evaluation of the Bambara pudding samples with different alternative cooking materials were generally acceptable (p<0.05) but pudding wrapped with banana leaf was significantly (p<0.05) rated low for colour and taste while others were comparable (p>0.05). In conclusion, bambara nut pudding cooked with alternative cooking materials contained polyethylene resins.

TU419 SETAC Human Health Risk Assessment Interest Group

B. Mulhearn, Ensafe Inc.

Fungicides - an overlooked compound group? Fate, effects, risk assessment and mitigation (P)

TU420 Ecological risk assessment of conazole fungicides in arable soils of the Czech Republic

J. Vacekova, M. Hvezdova, Masaryk University / Research Centre for Toxic Compounds in the Environment (RECETOX); P. Kosubova, Central Institute for Ecological Risk Assessment and Mitigation (P)

For regulatory purposes, noncarcinogens [1]. Presence of such compounds in arable soils represents potential risks of conazole fungicides in arable soils of the Czech Republic acquired in early 2015 [2]. In this study, 51 currently used pesticides and 9 transformation products were analysed by multi-residue pesticide analysis on LC-MS/MS after soil QuEChERS extraction. The data indicated that over 70% of soils contained at least one CF and the total concentration of CFs exceeded 0.01 mg/kg in 53% of soils. Epoxiconazole and tebuconazole also frequently exceeded 0.01 mg/kg (in 25% and 14% of soils respectively). The concentrations of CFs were epoxiconazole (48% of soils) and tebuconazole (36%), followed by fludioxonil (23%), prochloraz (2%), propiconazole (13%), cyproconazole (8%) and difenoconazole (7%). Overall, the CF fungicides are of environmental concern because they exceeded risk based thresholds, tend to form long-term residues in soil and rank among suspected carcinogens and endocrine disruptors. [1] EU pesticide database: ec.europa.eu/food/plant/pesticides/eu-pesticidesdatabase; [2] M. Hvezdova, et al., Sci. Total. Environ., vol. 613–614, pp. 361–370, 2018.

TU421 Concentration- and time-dependent dissipation, partitioning and plant accumulation of selected fungicides, insecticide, herbicide and transformation products in sand and soil

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In this study, the dissipation and partitioning dynamics and the extent of biotopkaue was measured and modeled for selected hazardous current-used fungicides (prochloroz, tebuconazole, fludioxonil, epoxiconazole), insecticide (chlorpyrifos), herbicide (pendimethalin) and for a transformation product (2-hydroxyatrazine) in agricultural soil and quartz sand as representatives of a real and a worst-case scenario. Dissipation, uptake to Lactuca sativa and the freely dissolved compounds was monitored along with the hydrophilic abnormally normalized sorption coefficients (Koc) were determined on day 12, 40, and 90 following the application of compounds at three fortification levels (0.1 - 1 - 0.1 mg/kg). Dissipation of tested compounds differed in soil and sand and was influenced by compound concentration. 2-hydroxyatrazine showed the longest persistence in soil among the tested compounds. The four fungicides showed very similar dissipation patterns and were more persistent in sand than in soil which implies that their main elimination mechanism in the environment is biodegradation. Plant roots were shown to accumulate higher amounts than shoots with root-to-shoot translocation factors (TFs) of 0.007-0.14 where the extent of root uptake was driven by compound partitioning. This was evidenced by the ability of Cmu to reliably (r² = 0.949) predict root uptake. Concentrations of non-target compounds did not exceed the maximum residue levels (MRLs) for lettuce. Koc values were in the range of literature values and were shown to increased (from 0 day to 40 day) as well as decreased for some compounds (from day 40 to 90 day) with time probably as a result of compound sequestration and competitive sorption, respectively. From the results, it follows that the tested compounds pose limited risks when presented in the soils for a given time. This was shown to be not persistent (except for 2-hydroxyatrazine), b) to accumulate in lettuce to extents below MRLs, c) to sorb effectively to soil, even in the presence of other co-solutes.

TU422 Evaluation of pesticides and fungicides transport using passive sampling devices in a vineyard catchment in South West France

J. Gaillard, Université de Bordeaux / EPOC UMR 5805; M. Dévier, University of Bordeaux / EPOC / LPTC UMR 5805 CNRS; K. Le Menach, P. Pardon, UMR CNRS EPOC Université Bordeaux / EPOC UMR 5805; G. DUPORTE, Université de Bordeaux / EPOC UMR 5805; F. Macary, Iristea Bordeaux; H. Budzinski, University of Bordeaux.

In vineyards of South West France, fungicides account for nearly 8 out of 10 pesticide applications which increases the likelihood of chronic exposure in adjacent ecosystems. The objective of the present study was to investigate the dynamics of pesticides and fungicides from agricultural drainage ditches to a third order stream in a vineyard catchment. Monitoring was a combination of passive and grab water sampling. Six sites with continuous-flowing-flow were monitored during a year using polar organic compounds integrative sampler (POCIS) retrieved monthly together with grab water samples collected monthly or bimonthly. Four sites with intermittent-flow were monitored using grab water samples only. Passive samplers such as POCIS enable the improvement of limits of quantification (LOQ) and estimation of time-weighted average concentrations over the exposure period for conservative compounds. To the best of our knowledge, 50 pesticides were targeted including 23 fungicides currently applied in the studied catchment. Extracted samples were analysed by liquid chromatography-tandem mass spectrometry (LC-MS/MS) and gas chromatography-tandem mass spectrometry (GC-MS/MS). Among the currently used pesticides in the catchment, preliminary results obtained from a sampling campaign conducted in spring allowed the quantification of 19 fungicides, 3 herbicides and 1 insecticide in passive samplers. In water samples, 9 fungicides and 1 insecticide were detected. Highest concentrations ( > 1 µg/g) were measured for the fungicides benalaxyl and dimethomorph. Fungicides such as cyproconz, kresoxim-methyl and iprodalcarb
were detected in passive samplers but were not detected in water samples suggesting the importance of combining sampling techniques to provide a more complete assessment of fungicide exposure in vineyard catchments.

**TU423**
Assessment of secondary exposure to fungicide residues in fruit-growing workers were assessed in arable crops in the Bordeaux region. G. DUPORTE, J. Gaillard, Université de Bordeaux / EPOC UMR 5805; E. Barron, University of Bordeaux, CNRS / EPOC UMR 5805; K. Le Menach, P. Pardon, UMR CNRS EPOC Université Bordeaux / EPOC UMR 5805; I. Baldi, University of Bordeaux / ISPED, EPIFENCE; F. Macary, Irstea Bordeaux; M. Déverie, University of Bordeaux / EPOC / LPTC UMR 5805 CNRS; H. Budzinski, University of Bordeaux. European Union countries have about 12 million agricultural holdings, and approximately 172 million hectares (39% of the total European land surface) of agricultural areas. Pesticides are used extensively in agricultural production to prevent pests, diseases, weeds or other plant pathogens to reduce yield losses and to guarantee a good harvest. In the recent decades, numerous studies have suggested adverse health effects associated to long-term pesticide exposure. Serious concerns have been raised about health risks resulting from occupational exposure. Nevertheless, the knowledge of occupational exposure levels and determinants to pesticides are still limited. The CANEPA project (Cancers and Exposures to Agricultural Pesticides) aims to characterise external contamination of agricultural workers and environmental contamination by pesticides in arboriculture. In this work we assessed the deaths of Korean victims whose exposure was formed in several apple holding, situated in south-west of France. Dislodgeable foliar residues (DFR) and pesticide residues on equipments or apples (wipe sampling) were studied during the different activities of apple growing (treatments, entry tasks, harvests). Atmospheric levels of pesticides (outdoor and indoor) were also determined using passive samplers (Polyurethane Foams, PUF) and low-volume sampler (in the trachea) with a high sensitivity (captain and dithianon), extensively used in apple growing, and their metabolites. High sensitive analytical methods were developed and validated, in this work, for the different collected samples based on gas or liquid chromatography coupled to hybrid high resolution mass spectrometry and to tandem mass spectrometry (LC/MS/MS). Levels of pesticide residues and source characterisation will be presented. These findings provide a better understanding of current practices and may help for reducing pesticide occupational exposure and health risks for fruit-growing workers.

**TU424**
Intra-tracheal administration of the disinfectant, chloromethylisothiazolinone/methylisothiazolinone (CMIT/MIT), in a mouse model to evaluate a causal association with death Y. Park, GLP Center, Catholic University of Daegu / APT; H. Kim, Graduate School of Medical Health Science, Catholic University of Daegu; B. Kang, Catholic University of Daegu, Graduate school of toxicity assessment G. Chaufan, University of Buenos Aires, Facultad de Ciencias Exactas y Naturales / Departamento de Química Biológica CONICET-UBA, Instituto de Química Biológica - Ciencias Exactas y Naturales (IQUIBICEN); C. Galvano, Universidad de Buenos Aires, Facultad de Ciencias Exactas y Naturales / Departamento de Química Biológica; M.D. Mudy, Universidad de Buenos Aires, Facultad de Ciencias Exactas y Naturales / Departamento de Ecología Genética y Evolución Facultad de Ciencias Exactas y Naturales; M. D. Rio de Molina, Universidad de Buenos Aires, Facultad de Ciencias Exactas y Naturales / Departamento de Química Biológica, CONICET-UBA, Instituto de Química Biológica - Ciencias Exactas y Naturales (IQUIBICEN); N. Andrioli, Universidad de Buenos Aires, Facultad de Ciencias Exactas y Naturales / Departamento de Ecología Genética y Evolución. The use of fungicides represents one of the most important factors in the control of pests and diseases, which affects the production systems of fruits and vegetables. It is known that most fungicide residues remain stable in food for long periods of time, increasing exposure risk for the general population. The aim of the present study was to evaluate the oxidative damage, the antioxidant response and the genotoxic effect in a human cell line (HeP-2) against the exposure of sublethal concentrations of the fungicide Iprodione. For this proposal, we determine the content of protein carbonyls as a marker of oxidative damage, the equivalent content of glutathione (GSH), the activity of antioxidant enzymes superoxide dismutase (SOD) and catalase (CAT) and from detoxifying enzyme GSH-S-transferase (GST), in 3 concentrations of Iprodione (1.5, 7 and 25 μg/ml). The cell division index, the replication index, the frequency of chromosomal aberrations and micronuclei were also determined in the presence of 7.5, 17.5 and 25 μg/ml of Iprodione. The L50 was determined using passive samplers (Polyurethane Foams, PUF) and low-volume sampler (in the trachea) with a high sensitivity (captain and dithianon), extensively used in apple growing, and their metabolites. High sensitive analytical methods were developed and validated, in this work, for the different collected samples based on gas or liquid chromatography coupled to hybrid high resolution mass spectrometry and to tandem mass spectrometry (LC/MS/MS). Levels of pesticide residues and source characterisation will be presented. These findings provide a better understanding of current practices and may help for reducing pesticide occupational exposure and health risks for fruit-growing workers.

**TU425**
Genotoxic response and alteration of intracellular redox balance in Hep-2 cell line by exposure to Iprodione G. Chaufan, University of Buenos Aires, Facultad de Ciencias Exactas y Naturales / Departamento de Química Biológica CONICET-UBA, Instituto de Química Biológica - Ciencias Exactas y Naturales (IQUIBICEN); C. Galvano, Universidad de Buenos Aires, Facultad de Ciencias Exactas y Naturales / Departamento de Química Biológica; M.D. Mudy, Universidad de Buenos Aires, Facultad de Ciencias Exactas y Naturales / Departamento de Ecología Genética y Evolución Facultad de Ciencias Exactas y Naturales; M. D. Rio de Molina, Universidad de Buenos Aires, Facultad de Ciencias Exactas y Naturales / Departamento de Química Biológica, CONICET-UBA, Instituto de Química Biológica - Ciencias Exactas y Naturales (IQUIBICEN); N. Andrioli, Universidad de Buenos Aires, Facultad de Ciencias Exactas y Naturales / Departamento de Ecología Genética y Evolución. The use of fungicides represents one of the most important factors in the control of pests and diseases, which affects the production systems of fruits and vegetables. It is known that most fungicide residues remain stable in food for long periods of time, increasing exposure risk for the general population. The aim of the present study was to evaluate the oxidative damage, the antioxidant response and the genotoxic effect in a human cell line (HeP-2) against the exposure of sublethal concentrations of the fungicide Iprodione. For this proposal, we determine the content of protein carbonyls as a marker of oxidative damage, the equivalent content of glutathione (GSH), the activity of antioxidant enzymes superoxide dismutase (SOD) and catalase (CAT) and from detoxifying enzyme GSH-S-transferase (GST), in 3 concentrations of Iprodione (1.5, 7 and 25 μg/ml). The cell division index, the replication index, the frequency of chromosomal aberrations and micronuclei were also determined in the presence of 7.5, 17.5 and 25 μg/ml of Iprodione. The L50 was determined using passive samplers (Polyurethane Foams, PUF) and low-volume sampler (in the trachea) with a high sensitivity (captain and dithianon), extensively used in apple growing, and their metabolites. High sensitive analytical methods were developed and validated, in this work, for the different collected samples based on gas or liquid chromatography coupled to hybrid high resolution mass spectrometry and to tandem mass spectrometry (LC/MS/MS). Levels of pesticide residues and source characterisation will be presented. These findings provide a better understanding of current practices and may help for reducing pesticide occupational exposure and health risks for fruit-growing workers.
The effects of salinity on chemical toxicity may warrant changes to future chemical assessments.

WE002
Photochemical transformation and intermediate formation processes in surface waters, in the context of climate change

D. Vione, M. Minella, C. Mineiro, University of Torino / Chemistry

Sunlight illumination of surface waters induces several photochemical reactions that play a role in the quality of water. The main factors involved in these reactions are the presence of sunlight, if any, and their indirect or sensitised transformation. In the latter case, sunlight is absorbed by naturally-occurring photostabilisers (e.g. chromophoric dissolved organic matter or CDOM), nitrate and nitrite that produce several photoproduction species that may undergo phototransformation reactions. The transients include, among others, the hydroxyl (·OH) and carbonate (CO$_3^{2-}$) radicals, singlet oxygen (O$_2^*$) and CDOM triplet states (CDOM$^*$). Their occurrence in surface-water environments is linked to irradiance and to key water parameters such as chemistry and depth [1,2]. The photoformation of dissolved compounds involves an interplay between molecular photoactivity and environmental features. Water chemistry and depth can affect both xenobiotics persistence and the possible formation of toxic or mutagenic intermediates. If a hazardous compound is preferentially produced by a certain photoreaction pathway, the environmental conditions can enhance or inhibit its formation in different surface-water environments [3]. The role of climate change on water chemistry and, as a consequence, on photochemical reactions is just starting to be investigated. The main difficulty is to disentangle climate effects from other disturbance factors (e.g. wastewater inputs) that may also operate and vary on the long term [4]. Climate change has the potential to deeply alter the photochemistry of freshwaters, but its effects could be very different in boreal versus temperate environments. In the former case the main effects would involve water light availability (e.g. tree line shifts, extended drought periods) would play key roles depending on the context. [1] Vione D, Minella M, Maurino V, Mineiro C. 2014. Chemistry Eur. J. 20:10590-10606. [2] Rosario-Ortiz FL, Canocona S. 2016. Environ. Sci. Technol. 50:12532-12547. [3] Avetta P, Fabbrì D, Minella M, Brigante M, Maurino V, Mineiro C, Pazzini M, Vione D. 2016. Water Res. 105:383-394 [4] Minelli L, Leoni B, Salmaso A, Savoie L, Sommaruga R, Vione D. 2016. Sci. Total. Environ. 541:247-256.

WE003
How Pharmaceutical Industrial waste can make your medicines ineffective

N. Verma, Babdi University of Emerging Sciences & Technology / Pharmacy

Spread over 380 square kilometres in Himachal Pradesh’s Solan district, the Babdi-Barotiwala-Nalagarh (BBN) industrial area is one of India’s largest pharmaceutical producing hubs. The region hosts around 500 small, medium and large pharma units and accounts for 35 per cent of Asia’s total medicine production. But rapid industrialisation and a lax attitude towards safe disposal and management of pharma waste has concerns about the effects of pollution on the environment and health. Liquid waste from these units is also discharged through pipes and other outlets that open behind the plant or run underground and open into bushy areas. This released wastewater accumulates in or flows through nallahs, canals and rivulets into the Sirsa river. Effluents are also injected into the ground at night by digging bore wells or released during rains. Due to such practices, the BBN region is referred to as a hotspot of antibiotic pollution. Many companies manufacture formulations, or finished products such as tablets and syrups. Some companies also manufacture active pharmaceutical ingredients (APIs) or the main biologically active ingredient used in formulations, including antibiotics. These APIs can enter the environment due to insufficient treatment or improper disposal of waste and weak environmental regulations. They are among environmental persistent pharmaceutical pollutants which have not degraded completely during treatment. They may influence the genetic makeup of bacteria, leading to the survival of resistant bacteria and spread of antimicrobial resistance (AMR), a public health threat. The result of our study showed that all gaps leading to the release of

Prioritisation and Intelligent Testing of Pharmaceuticals in the Environment (P)

WE001
Development of a modelling framework for estimating the sorption of pharmaceuticals in soils

L. Carter, University of York / Environment Department; J. Wilkinson, The University of York / Natural and Built Environments; A. Boxall, University of York / Environment Department

Ionisable pharmaceuticals comprise a significant and increasing proportion of chemicals used in Europe. At typical environmental pH, ionisable pharmaceuticals can become charged. Speciation can alter the fate and behaviour of a chemical in the environment including its sorption potential to soils and sludge. It is essential that the behaviour is recognised within chemical risk assessment and predictive approaches are able to account for how speciation alters chemical sorption. Several authors have proposed approaches to predict the sorption of ionisable chemicals in soils. However, these models are typically based on training sets containing a multitude of organic chemicals and their ability to predict ionisable pharmaceutical sorption specifically needs to be evaluated. We therefore evaluated a range of predictive approaches, that take into account sorbent properties (i.e. soil characteristics), for their suitability for estimating sorption of pharmaceuticals in soil. The evaluations were done using a database of high quality experimentally-determined pharmaceutical sorption coefficients provided by industry partners. Models developed for specific classes of ionisable chemicals (i.e. cations or anions) performed better in comparison to simple generic models, which assume that hydrophobicity is the key sorption mechanism and neglect to take into account of the effects of chemical speciation. Nevertheless, model predictions for ionisable pharmaceuticals still performed poorly ($r^2 < 0.5$). Sorption coefficients for organic cations were typically within an order of magnitude of experimental values where position was consistent with the charge of sorption on organic matter and to clay minerals. As sorption of neutral and anionic compounds were not well explained by the evaluated models, further model development was required for adequate prediction of soil sorption coefficients for these classes of molecule. A decision tree framework to guide the selection of appropriate sorption models by taking into account soil pH and ionisable functional groups has been created. This incorporates previously published models that performed well in our analysis and the development of new sorption models. Work is currently on-going to review sludge sorption models and will be presented. The authors acknowledge EU/EFPIA Innovative Medicines Initiative Joint Undertaking (iPIE grant n° 115735) for the financial support.

TU428
From mother to offspring: multigenerational effects of carbendazim at individual and subcellular levels in Daphnia magna

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Anthropogenic activities such as the use of pesticides may have indirect disastrous consequences on species such as Daphnia magna, which has a high potential to end up in aquatic ecosystems mainly through runoff. The deleterious effects observed at the population level can often be depicted or explained by changes in homeostasis at cellular and individual levels. In the present study, an isoclonal population of Daphnia magna (clone k6) was exposed to an environmentally relevant concentration (5 µg/L) of carbendazim during three generations. The effects of carbendazim on survival/longevity, biochemical biomarkers (cholinesterase, catalase and glutathione S-transferase), lipid peroxidation and energy-related parameters (carbohydrates, lipids and proteins jointly with energy available and energy consumption) were assessed in some generations. The long-term exposure to carbendazim presented no effect on the intrinsic rate of natural increase ($r$) and length of adult D. magna. However, daphnids longevity decreased at F12 generation and an increase in DNA damage from generation F3 to F13 was found when compared to daphnids in clean medium. Cholinesterases and glutathione S-transferase activities and lipid peroxidation showed differences between non-exposed and exposed populations to carbendazim. However, for catalase and energy-related parameters (except lipids) no differences were observed between these two Daphnia populations. Overall, at the tested concentration, carbendazim induced low effects under a long-term exposure to a daphnid population.
APIs in the environment are plugged. The discharge of pharmaceutical effluents should take place through proper waste management techniques and stringent environmental regulations. Currently, effluent standards are limited to chemical contaminants such as heavy metals. The government must adopt a new AMR-centric approach of waste management which considers APIs as a chemical contaminant. Laws must be made to ensure that there are no APIs in treated effluents. The government should support small-scale manufacturers to install and implement environmentally sound waste treatment and disposal techniques. Manufacturers with high-end WWTPs should also be strictly monitored. The SPCBs should conduct surveillance of APIs or antibiotic residues in the treated effluents and make data publicly available.

**WE004**
The environmental concentration and evaluation of active ingredients in pharmaceuticals in rivers flowing through urban area in Japan

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The active ingredients in pharmaceuticals are discharged into the aquatic environment after use mainly through sewage treatment facilities. There is concern about adverse effects on wildlife due to the possibility of the presence of highly susceptible species. However, we still know little about what kind of impact on wildlife in the environment. Based on these backgrounds, we set up environmental impact assessment methods for the pharmaceuticals according to AMED’s strategy and evaluated the risks. We have measured the environmental concentrations (MEC) of 31 kinds of active ingredients in marketing medicine, using liquid-chromatography with mass spectrometry, in representative seven urban rivers in Japan, once every four seasons in 2015-2016. The maximum detected concentrations of the active ingredients exceeded 100ng/L are shown below: olmesartan (571ng/L), valsartan (494ng/L), candesartan (113ng/L), losartan (117ng/L) for antihypertensive agents, and sulpiride (546ng/L) for antipsychotic agent, crotamiton (445ng/L) for antibacterial agent, ketoprofen (150ng/L) for analgesic antiprretic agent, bezafibrate (200ng/L) for hyperlipidemia treatment drug, crotamiton (445ng/L) for antipsychotic agent. Among target ingredients, the detect concentration of active ingredient contained in pharmaceuticals for the lifestyle-related disease, hypertension and lipid metabolism related disease, tended to be higher. The concentrations in the winter or spring was observed a higher tendency, but the detected concentrations of active ingredients greatly varied depending on river according to the type of lifestyle and the type of pharmaceuticals been spread. It was indicated that the detected active ingredients were derived mainly from sewage treatment water as it depends on the concentration of sucrose measured at the same time. The pharmaceuticals whose maximum detected concentrations of active ingredients in each river water exceeded the predictive environmental concentrations (PEC) were five ingredients of candesartan, olmesartan, lorazepam, rosuvastatin and epinastine, even when the dilution ratio was doubled. This result suggests that in some circumstances it is necessary to consider compartment for dilution in the environment which sets it. Regarding the health effects on humans, the actual concentration for each daily minimum dose for each pharmaceutical ingredient was from 0.086% of lorazepam to 0.000001% of chlorbic acid.

**WE005**
Evaluation of simple exposure models used for environmental prioritisation of active pharmaceutical ingredients

J. Wilkinson, The University of York / Natural and Built Environments; A. Boxall, L. Carter, University of York / Environment Department; E. Burns, University of York

Targeted quantification using analytical methods such as high performance liquid chromatography followed by tandem mass spectrometry (HPLC-MS/MS) are effectively used to monitor trace-levels (ng/L) of active pharmaceutical ingredients (API) in the aquatic environment. However, as more than 1500 chemicals are currently in-use as pharmaceuticals, the high cost of HPLC-MS/MS prohibits its widespread use in the monitoring and prioritisation of APIs. Predictive exposure models can offer cheaper treatment water as it depends on the concentration of sucrose measured at the same time. The pharmaceuticals whose maximum detected concentrations of active ingredients in each river water exceeded the predictive environmental concentrations (PEC) were five ingredients of candesartan, olmesartan, lorazepam, rosuvastatin and epinastine, even when the dilution ratio was doubled. This result suggests that in some circumstances it is necessary to consider compartment for dilution in the environment which sets it. Regarding the health effects on humans, the actual concentration for each daily minimum dose for each pharmaceutical ingredient was from 0.086% of lorazepam to 0.000001% of chlorbic acid.

**WE006**
The role of the water-sediment simulation test and its outcome in the environmental risk assessment (ERA) of pharmaceuticals

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In view of the revision of the “Guideline on the environmental risk assessment of medicinal products for human use” (EMEA/CHMP/SWP/4447/00, June 2006, rev. 2) current fate assessment is highly discussed concerning the water-sediment simulation test in tier A of the guideline. At the moment, only the partitioning to sediment triggers further assessment in the next tier of the guideline. Derived half lives were not used in refined predicted exposure concentration (PEC) calculations and transformation products (TP) were not specifically considered in the ERA. Now, UBA evaluates the data received in the last 10 years in order to determine the gaps and the benefit of the current fate assessment for the overall risk assessment e.g. classification of persistence. Especially the role of TP in the environment due to their frequent higher mobility compared to the parent compound is considered in the presented research. As a first step an overview is prepared on the overall persistence of pharmaceuticals in the environment in the last 10 years. It is clearly demonstrated that total system half-lives already show a high persistence of pharmaceuticals in the aquatic environment. Furthermore it should be considered that especially for the sediment compartment often no kinetic model fits well enough to predict DT50 values. The risk of ground water contamination by bank filtration will be estimated by the use of physio-chemical properties e.g. lipophilicity / hydrophilicity by comparing parent compound and TP. The occurrence and identification of TP is often appeared to be something which is only “nice to have” but not really relevant for the risk assessment of human pharmaceuticals. TP are often more polar and stable in environmental compartments than the parent compounds. This is of high relevance for groundwater contamination. The identification strategy of relevant TP is still often missing in provided studies. The water sediment simulation study is the only experimental study in the ERA which gives information about the possible behaviour and occurrence of parent compounds and TP in surface waters. Such results are relevant for monitoring and for the understanding which compartments are affected by pharmaceuticals. Instead of waiting OECD 308, the results should be better included in the ERA and communicated.

**WE007**
Expert System to Inform BCF Testing Strategies for Pharmaceuticals

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An important part of the environmental risk assessment (ERA) of pharmaceutical ingredients is the identification of the persistent, toxic and bioaccumulative properties (PBT assessment) of the molecule. Regulations and guidelines on how to conduct this assessment and what empirical data are required to do so are increasingly complex. Currently a large number of fish are used as part of the ERA process, particularly for experiments to determine the bioconcentration factor (BCF), even though research developments and guidelines already contain opportunities to significantly reduce the number of fish used via alternative methods. The identification of relevant TP is still often missing in provided studies. The water sediment simulation study is the only experimental study in the ERA which gives information about the possible behaviour and occurrence of parent compounds and TP in surface waters. Such results are relevant for monitoring and for the understanding which compartments are affected by pharmaceuticals. Instead of waiting OECD 308, the results should be better included in the ERA and communicated.
Effects of duloxetine and econazole on freshwater species towards individual and combined conditions
G AMARIELI, Universidad de Alcalá; K. Boltes, University of Alcalá / Chemical Engineering; J. Valim"ah-Traverso, M. García, P. Letón, M. Marina, R. Rosal, University of Alcalá

Thousands of biologically active pharmaceutical ingredients (APIs) are used in human medicine and veterinary medicine worldwide. Nowadays, the occurrence of pharmaceuticals in aquatic environments is a well-established issue, where there are still gaps in our knowledge on the fate and effects of these compounds in the environment. Evaluating API ecotoxicology is even more challenging due to uncertainties about appropriate dosages, durations of exposure, range of sensitive taxa, sensitivity of developmental stages, and toxicological endpoints. More attention should be paid on the non-target organisms and the chiral nature of contaminants. This work assess the toxicity of the antidepressant drug Duloxetine and the antifungal Econazole, individually and combined, on three freshwater species—algae, crustacean and duckweed, using APIs concentration from 0.039 to 100 mg L\(^{-1}\). Level 1 type of drugs interactions were determined using the Combination Index-isobologram method. The enantiomers concentration of the target compounds in the culture media were 440 ppm. COX and succeeded to detect the inhibitory activity of antidepressants in SEs. Inhibition of antidepressants in wastewater treatment plants (WWTPs) could be a rapid assessment of the risk of antidepressants to aquatic organisms. Within the first six months of the study, mides showed to be less sensitive to carbamazepine after long-term low-level exposure. Chronic toxicity tests to study the authors federal by the Federal Ministry of Education and Research (BMBF) for funding (NiddaMan, project support code: 02WRM1367A).

WE008

Development of a quantitative Adverse Outcome Pathway-informed model to predict the risk posed by mixtures of non-steroidal anti-inflammatory drugs to fish
P Marmon, J. Kayode, Brunel University London / College of Life Sciences; S. Owen, AstraZeneca / Safety Health Environment; L. Margiotta-Casaluci, Brunel University London / Institute of Environment, Health and Societies

The presence of low concentrations of non-steroidal anti-inflammatory drugs (NSAIDs) in the aquatic environment has raised the concern that chronic exposure to these compounds may cause adverse effects in wild fish populations, similar to those observed in human patients. This potential scenario has led to the inclusion of diclofenac in the European Union Watch List of emerging pollutants. Although the effects of diclofenac in fish have been investigated in over twenty published studies to date, the complexity of NSAIDs toxicity is such that many uncertainties still exist about the significance of those findings for environmental risk assessment. We hypothesise that the perturbation of cyclooxygenase (COX) activity in healthy fish tissues is the leading cause of adverse effects, as it is in humans. On the basis of this mechanistic starting point, we propose a quantitative Adverse Outcome Pathway-informed (AOP-G) approach to predict the ecopharmaceutical and pharmacodynamic aspects of NSAIDs toxicity. After extracting all NSAIDs toxicity data available in the scientific literature, we applied drug uptake models to predict the plasma concentrations at which different effects would occur in laboratory studies. As all NSAIDs act by inhibiting the enzymes COX1 and/or COX2, we applied mixture pharmacology approaches to express the plasma effect concentrations as minimal effect doses (MEDs) and effective concentrations as dichlorofenac equivalents. A similar approach was used for measured and predicted river concentrations of NSAIDs, which were used to predict plasma concentrations of NSAIDs in wild fish. The overlay of the two approaches led to a visual model that enables a rapid assessment of the risk posed by environmental levels of NSAIDs to trigger multi-scale adverse effects. The major strengths of the model are the ability to express the toxic potential of NSAIDs mixtures, expressed as dichlorofenac-equivalents. We propose that this mechanistic approach may provide a useful predictive tool to support the implementation of effective NSAIDs ecocarcinogenicity strategies and facilitate the regulatory interpretation of past and future toxicity data.

WE009

Evolution in the lab - How can we study the chronic exposure to pharmaceuticals over multiple generations?
K Heye, Goethe University Frankfurt/ Main / Aquatic Toxicology; A. Schmidt, Goethe University Frankfurt / Aquatic Ecotoxicology; J. Oehlemann, Johann Wolfgang Goethe-Universitat Frankfurt / Aquatic Ecotoxicology

Larval exposure of non-target organisms is poorly covered in the assessment of chemicals. Especially pharmaceuticals play an important role when it comes to long-term exposure. They enter the environment throughout the year and therefore pose a continuous risk to organisms. One of these ubiquitously detected pharmaceuticals is the antiepileptic drug carbamazepine (CBZ). Hardly degraded during conventional wastewater treatment, it contaminates a majority of water bodies. CBZ was identified in WWTPs as well as dichlorofenac as dichlorofenac equivalents. A similar approach was used for measured and predicted river concentrations of NSAIDs, which were used to predict plasma concentrations of NSAIDs in wild fish. The overlay of the two approaches led to a visual model that enables a rapid assessment of the risk posed by environmental levels of NSAIDs to trigger multi-scale adverse effects. The major strengths of the model are the ability to express the toxic potential of NSAIDs mixtures, expressed as dichlorofenac-equivalents. We propose that this mechanistic approach may provide a useful predictive tool to support the implementation of effective NSAIDs ecocarcinogenicity strategies and facilitate the regulatory interpretation of past and future toxicity data.
The ubiquity of pharmaceutical compounds in the aquatic environment, their growing use and their potential to elicit biological effects even at low concentrations is a major concern to environmental health and safety management. In recent years an increasing number of studies have addressed the presence and toxicity of various pharmaceuticals, using various biological endpoints in different biological models, and reporting varying effects. Accordingly, a systematic quantitative assessment is key to improve current understanding of the ecological risks of pharmaceuticals to non-target organisms in the aquatic environment. To unravel patterns in biological responses across aquatic taxa a meta-analysis was performed on reported effects of exposure to pharmaceutical compounds (according to therapeutic class). Minimum response concentration and biological responses were collected from selected studies based on a set of objective criteria considering organisms’ exposure to pharmaceuticals under controlled conditions. For a response sensitivity analysis various endpoints were considered, namely biochemical, developmental (e.g. growth), reproductive and behavioral responses, as well as lethality, in studies reporting effects on aquatic taxa. The comparative sensitivity analysis of biochemical endpoints highlighted the sensitivity of molecular responses, followed by individual level-responses (e.g. behavior and growth), yet variable sensitivity depending on among taxa were observed when considering different pharmaceutical classes. Data availability and comparability limited qualitative analysis, yet the meta-analysis provided a key framework to compare effects influence on development and reproduction of crustacean and fish exposed to pharmaceuticals. Overall the implications of current findings for environmental monitoring and ecological risks of pharmaceuticals in aquatic ecosystems are discussed.

WE015
Leveraging Pharmacological Data for Prioritization of the Ecological Risks of Chiral Pharmaceuticals
E. Umangayav, University of California, Riverside / Department of Environmental Sciences
Pharmaceuticals are frequently detected in wastewater and the environment at concentrations ranging from ng/L to µg/L. More than 50% pharmaceuticals are chiral compounds. Enantiomers of chiral pharmaceuticals have been shown to exhibit differences in environmental fate, transport and toxicity. Since more than 2,500 pharmaceuticals are currently in use, it is implausible to carry out whole organism toxicity studies of all pharmaceuticals. However, there is a wealth of knowledge available from drug discovery and development research that can be leveraged for predicting potential environmental exposure and effects of chiral pharmaceuticals. Assuming evolutionary conservation of primary drug target, read-across method can be used to predict the potential effect of chiral pharmaceuticals. In this study, we estimated the stereoselective effect of 11 chiral pharmaceuticals using the fish plasma model. We found metoprolol had high risk with an effect ratio, ER (ratio of human therapeutic plasma concentration to fish plasma concentration at steady state) that was less than 1.0, whereas propranolol, salbutamol, fluoxetine and venlafaxine were medium risk (1.0 < ER < 30). However, stereoselectivity was predicted in all compounds except atenolol and pindolol. In this study, we showed the fish plasma model has considerable potential as a predictor of stereoselective toxicity of chiral pharmaceuticals.

WE014
Effects of benzoylcoenzyme exposure at different levels of the biological hierarchy on Daphnia magna
M. Parolini, University of Milan / Department of Environmental Science and Policy; B. De Felice, Università degli Studi di Milano; C. Ferrario, University of Milano Bicocca; N. Salgueiro, University of California, Riverside / Department of Environmental Sciences
A number of monitoring studies have shown that benzoylcoenzyme (BE), a metabolite of cocaine, is the main illicit drug residue measured in both wastewater and surface waters worldwide. Although the aquatic concentration of BE can be considered still low, the exposure to this molecule may cause diverse adverse effects. The few studies that have addressed this topic to date investigated the toxicity of this molecule towards invertebrate and vertebrate aquatic non-target organisms have shown different detrimental effects at low levels of the biological organization, mainly at biochemical, molecular and cellular levels. However, to date no one study has evaluated the consequences of BE exposure to the higher levels of ecological hierarchy. Thus, the present study was aimed at investigating the toxicity of a 48-h exposure of 6 concentrations of BE, similar to those found in aquatic ecosystems (0.5 µg/L and 1.0 µg/L), on the cladoceran Daphnia magna at different levels of the ecological hierarchy. We relied on a multi-level approach focusing on the effects at biochemical/biomolecular (biomarkers), individual (swimming activity) and population (reproduction) levels. As previous studies of BE have shown that this molecule can induce oxidative stress, we assessed the amount of reactive oxygen species and of the activity of antioxidant (SOD, CAT, and GPX) and detoxifying (GST) enzymes and the lipid peroxidation (TBARS) as oxidative stress endpoints. We also measured the acetycholinesterase (AChE) activity because this enzyme is strictly related to behavioral changes in aquatic organisms. Alterations in the swimming behaviour of D. magna were investigated by a video tracking analysis, while the consequences on the reproduction were assessed by a chronic toxicity test. Our results showed that the exposure to two BE concentrations similar to those found in aquatic ecosystems induced oxidative stress and inhibited the activity of AChE, affecting the swimming behavior and the reproduction of Daphnia magna individuals.

WE015
Impact of the antibiotic drug metformin and its transformation product guany lurea on brown trout (Salmo trutta f. fario)
S. Jacob, Universität Tübingen / Animal Physiological Ecology; L. Kundy, M. Biecker, University of Tübingen; R. Trischkorn, University of Tübingen / Animal Physiological Ecology
The last decades, the number of patients suffering from diabetes type 2 is increasing. Consequently, a rising consumption of antibiotic drugs as metformin (MF) has become obvious. In wastewater treatment plants, metformin is only partially retained and also transformed to guany lurea (GU) leading to high concentrations of both compounds in surface waters. However, possible effects of MF and GU in aquatic organisms are far from being understood. The aim of this study is therefore to investigate influences of MF and GU on different metabolic pathways and behaviour in different life stages of brown trout (Salmo trutta f. fario). Juvenile trout (age: 8 month) were exposed for 4 weeks at 7°C to different concentrations of MF (0, 1, 10, 1000 µg/L) and GU (0, 1, 10, 100, 1000 µg/L). Additionally, eggs of brown trout in the eyed ova stage were exposed to different metformin concentrations (0, 1, 10, 100, 1000 µg/L) at 7°C & 11°C (regarding possible interactions of chemical toxicity & temperature). To show influences on the embryo development, mortality, hatching rate, and heart rate were recorded. Tissue samples were taken three and eight weeks after the end of the sac-fry stage. In all experiments, several endpoints characterizing fish health were investigated, including the histological condition of the liver, alterations in the stress protein level (HSP70), changes in the intestinal microbiome and additionally the glycogen storage in the liver of MF-exposed fish. Besides, swimming and predator-prey behaviour were investigated. There was no influence of MF on the developmental parameters in brown trout larvae. Neither behaviour nor stress protein level were influenced by MF. The liver tissue of the MF-exposed trout was in a good condition. The glycogen storage was tendentially increasing in MF-exposed fish compared to the control, whereas the glycogen content of the trout exposed to 1000 µg/L MF was partially decreased. The intestinal microbiome of MF-exposed larvae showed a significantly different composition compared to the control. The results for the experiment with GU will be presented (analyses not yet finished). This work is part of the project Eff-Net (Effect Network in Water Research) funded by the German Federal Environmental Agency’s Water Network Baden-Württemberg. In this study the effects of both MF and GU on the fish were compared and will be discussed. Possible implications of these differences are going to be discussed and discussed.

WE016
Effect of life-cycle exposure to environmentally relevant concentrations of metformin and its metabolite guany lurea on F1 progeny 28 days post hatch.
Z.P. Pandelides, University of Ontario Institute of Technology; E. Ussery, University of Ontario Institute of Technology / Biological Sciences; J. Guchardi, University of Ontario Institute of Technology / Faculty of Science Aquatic Toxicology; D.A. Holdway, University of Ontario Inst. of Tech / Science Research Studies have demonstrated that the type 2 diabetic drug metformin and its only known metabolite, guany lurea, are common environmental contaminants found in the ng-µg/L concentration range in surface waters and wastewater effluent. This should be of concern as recent work in our lab shows that Japanese medaka (Oryzias latipes) exposed to environmentally relevant concentrations of metformin and guany lurea from embryo through 28 days post-hatch have a significant decrease in length and weight of both males and females when compared to control fish. Furthermore, our studies show that larvae exposed for 28 days to both compounds have a significant dysregulation in lipid and fatty acid metabolism, possibly leading to this stunted growth. A full life-cycle exposure to both compounds at environmentally relevant concentrations, alone and in combination, was administered in order to examine the effects of chronic exposure to the F1 progeny. The effects of metformin and guany lurea on the length and wet weight were compared 28 days post hatch and will be discussed. Possible implications of exposure to metabolomics and gene expression will be explored.

WE017
Life-cycle effects in Oryzias latipes exposed to environmentally relevant concentrations of metformin and its metabolite, guany lurea.
E. Ussery, University of Ontario Institute of Technology / Biological Sciences; Z.P. Pandelides, University of Ontario Institute of Technology; J. Guchardi, University of Ontario Institute of Technology / Faculty of Science Aquatic Toxicology; D.A. Holdway, University of Ontario Inst. of Tech / Science Research Studies have demonstrated that the type 2 diabetic drug metformin and its only known metabolite, guany lurea, are common environmental contaminants found in the ng-µg/L concentration range in surface waters and wastewater effluent. As the majority (>90%) of metformin is metabolized into metformin hydroxyurea (MF) has become obvious. In wastewater treatment plants, metformin is only partially retained and also transformed to guany lurea (GU) leading to high concentrations of both compounds in surface waters. However, possible effects of MF and GU in aquatic organisms are far from being understood. The aim of this study is therefore to investigate influences of MF and GU on different metabolic pathways and behaviour in different life stages of brown trout (Salmo trutta f. fario). Juvenile trout (age: 8 month) were exposed for 4 weeks at 7°C to different concentrations of MF (0, 1, 10, 1000 µg/L) and GU (0, 1, 10, 100, 1000 µg/L). Additionally, eggs of brown trout in the eyed ova stage were exposed to different metformin concentrations (0, 1, 10, 100, 1000 µg/L) at 7°C & 11°C (regarding possible interactions of chemical toxicity & temperature). To show influences on the embryo development, mortality, hatching rate, and heart rate were recorded. Tissue samples were taken three and eight weeks after the end of the sac-fry stage. In all experiments, several endpoints characterizing fish health were investigated, including the histological condition of the liver, alterations in the stress protein level (HSP70), changes in the intestinal microbiome and additionally the glycogen storage in the liver of MF-exposed fish. Besides, swimming and predator-prey behaviour were investigated. There was no influence of MF on the developmental parameters in brown trout larvae. Neither behaviour nor stress protein level were influenced by MF. The liver tissue of the MF-exposed trout was in a good condition. The glycogen storage was tendentially increasing in MF-exposed fish compared to the control, whereas the glycogen content of the trout exposed to 1000 µg/L MF was partially decreased. The intestinal microbiome of MF-exposed larvae showed a significantly different composition compared to the control. The results for the experiment with GU will be presented (analyses not yet finished). This work is part of the project Eff-Net (Effect Network in Water Research) funded by the German Federal Environmental Agency’s Water Network Baden-Württemberg. In this study the effects of both MF and GU on the fish were compared and will be discussed. Possible implications of these differences are going to be discussed and discussed.
guanuylurea during the waste water treatment process, it is found in the environment in higher concentrations than metformin, usually in the µg/L concentration range in surface waters. This is concerning, as our recent research shows that Japanese medaka (Oryzias latipes) exposed to environmentally relevant concentrations of metformin (1.0-100 µg/L) and guanuylurea (1.0-100 ng/L) from embryo through 28 days post hatch have a significant decrease in length (mm) and weight (mg) of both male and female offspring compared to control fish, with guanuylurea appearing to be roughly 1,000 times more potent than its parent compound, metformin. Furthermore, these studies show significant changes in the metabolome of 28 day old male medaka exposed to both metformin and guanuylurea, indicating significant dysregulation in fatty acid and lipid metabolism. These results raise concern regarding the consequences of a full life cycle exposure, including the important reproductive phase. Thus, a full life-cycle continuous exposure experiment was undertaken utilizing both compounds at environmentally relevant concentrations, and in combination, was conducted in order to examine the effects of chronic exposure on growth, reproduction, steroid production, and gene expression. Results will be discussed.

WE018 Environmental Fate and Effects of the Antidiabetic Drug Metformin and Its Transformation Product Guanuylurea J. Straub, F.Hoffmann-La Roche Ltd / Roche Group Safety, Health & Environmental Protection; D.J. Caldwell, Johnson & Johnson / Environment Health Safety Sustainability; V. D'Acio, Quantum Management Group, Inc.; T. Davidson, Bristol-Myer Squibb / EHS; K. Kappler, Johnson & Johnson; S. Owen, Bristol-Myer Squibb / EHS; B. De Felice, Universidade de Lisboa / MARE and AstraZeneca / Safety Health Environment; B. Simon, Bristol-Myer Squibb / EHS; Hettich, Merck KGaA; J.G. Tell, Merck & Co., Inc. / Global Safety the Environment Metformin (MET) is an active pharmaceutical ingredient (API) with very high patient use worldwide that is excreted in unchanged form. This has led to concerns about the potential aquatic life impacts associated with the presence of MET in surface water. Guanuylurea (GUU) is formed from the transformation of MET by WWTP, and both MET and GUU are further degraded in the environment. A comprehensive aquatic life risk assessment of MET and GUU in surface water is presented that is based on literature data, previously unpublished data from industry studies conducted to support new drug registration applications as well as new studies commissioned to fill data gaps. Predicted environmental concentrations (PECs) for MET were modelled based on documented usage for the USA with the PhATE model and for the European Union with the GREAT-ER model. These PECs were compared with measured environmental concentrations (MECs) for both the USA and EU. A predicted no effect concentration (PNEC) for MET was derived by deterministic procedures based on multiple chronic studies with algae (4), daphnids (5) and fish (5, two species). Both the PEC/PNEC and MET/GUU risk characterization ratios were well below 1, indicating no significant risk for MET with high Margins of Safety. However, since MET is known to be primarily degraded during wastewater treatment to GUU, relevant chronic studies for GUU were conducted to derive a PNEC. In addition, PECs were derived for GUU for the USA and EU as above for MET. Fate and removal/in-stream-loss parameters for both MET and GUU show good alignment. PECs were derived in the USA and Europe. The PEC/PNEC and MEC/PNEC risk characterization ratios for GUU were also below 1. We conclude there is no significant risk to aquatic life for both MET and its transformation product GUU.

WE019 Fluoxetine exposure modulated antioxidant and anxiety-related gene expression altering swimming activity in zebrafish embryos B. De Felice, Università degli Studi di Milano; A. Ghilardi, L. Del Giacco, University of Milan; M. Parolini, University of Milan / Department of Environmental Science and Policy The massive release of human pharmaceuticals into the aquatic ecosystems continues to be a serious environmental problem. Although pharmaceuticals, psychotropic drugs such as antidepressants are one of the main therapeutic classes detected in freshwater worldwide. Selective serotonin reuptake inhibitors (SSRIs) are commonly the first-line antidepressant drugs prescribed to alleviate disorder in humans, and fluoxetine (FLX), the active principle of the Prozac, is one of the most prescribed worldwide. FLX enters aquatic ecosystems, whereby it has been detected in high ng/L to low µg/L concentrations. Therefore, this study was aimed at investigating 1) the effects induced by two concentrations of FLX (50 ng/L and 500 ng/L) on behavior and 2) if changes in the expression of neurotoxicity-related genes were related to changes in the swimming behavior of zebrafish (Danio rerio) embryos at 96 hours post fertilization (hpf). Our results showed that FLX exposure upregulated sod1, cat and gac, suggesting that this drug can induce an overproduction of pro-oxidant molecules. In addition, changes in the expression of slc6a3, slc6a4a, slc6a4b, slc6a11 and 2) if changes in the expression of neurotoxicity-related genes were related to changes in the swimming behavior of zebrafish (Danio rerio) embryos at 96 hours post fertilization (hpf). Our results showed that FLX exposure upregulated sod1, cat and gac, suggesting that this drug can induce an overproduction of pro-oxidant molecules. In addition, changes in the expression of slc6a3, slc6a4a, slc6a4b, slc6a11 genes indicated that FLX can affect neurotransmission and, consequently, alter swimming behavior of embryos, as demonstrated by the significant reduction of the distance moved by treated embryos in response to an external stimulus.

WE020 Bio-Optical probing of Beazafibrate toxicity in model marine diatom Phaeodactylyum tricornutum B. Daudé, MARE - Marine and Environmental Sciences Centre / Centro de Oceanografia; A. Matos, BioSLIBiosystems and Integrative Sciences Institute / Plant Functional Genomics Group, T. Cabrilla, IPMA IP; J.C. Marques, Universidade de Coimbra / MARE, Dep. of Life Sciences, Coimbra University; I. Caçador, Faculdade de Ciências da Universidade de Lisboa / MARE - Marine and Environmental Sciences Centre; P. Reis-Santos, MARE - Marine and Environmental Sciences Centre; H.N. Cabral, Faculdade de Ciências da Universidade de Lisboa / MARE-FUCUL; V.F. Fonseca, MARE Marine and Environmental Sciences Centre The occurrence and fate of pharmaceutical active compounds in aquatic environments has become a major cause for concern due to their effects on humans and aquatic ecosystems. The high consumption of blood lipid regulators is leading to increased occurrence of fibrates in natural streams and wastewater effluents. Fibrates are a class of drugs derived from fibrin acid widely used to reduce plasma triglycerides and raise the level of high-density lipoprotein cholesterol. Specifically, Beazafibrate is extensively used as a lipid regulator with consumption greatly increasing over the years in developed countries. Due to its large use and its persistence, beazafibrate has been detected in surface and drinking waters as well as in wastewater effluents. This has led to serious concerns regarding its molecular mechanisms of action and the relationship with organism fitness. Therefore, this study was conducted in order to examine the effects of chronic exposure on growth, reproduction, steroid production, and gene expression. Results will be discussed.

WE021 Environmental Risk Assessment for the Active Pharmaceutical Ingredient Mycofenolic Acid in European Surface Waters B. Daudé, F.Hoffmann-La Roche Ltd / Roche Group Safety, Health & Environmental Protection; A. Haener, F. Hoffmann-La Roche Ltd / Group SHE (LSO) An Environmental Risk Assessment (ERA) was performed for the active pharmaceutical ingredient mycofenolic acid (MPA) for Europe. MPA is an older immune inhibitor developed in the USA in the 1990s by Syntax, Inc., now a part of the Roche Group. So far, no sufficient dataset describing biodegradability, environmental fate or ecotoxicity for MPA nor an ERA for MPA have been available. The present ERA is based on old environmental data from Syntax and on new tests, all performed under GLP quality assurance, for physico-chemical characteristics, partitioning, environmental fate, biodegradability and (sub)chronic/chronotoxicity and on sales amounts for the products containing MPA in Europe. A predicted environmental concentration (PEC) for MPA was calculated based on compound actual use data from IMS Health, Inc. per annum and country, incorporating population data from Eurostat, for the decade 2004-2014. A crude initial PEC was derived based on standard ERA assumptions of no removal in sewage treatment or surface waters. The crude PEC was refined by incorporating predicted sewage works removal, based on new biodegradability data, and by country-specific dilution factors. The lowest of the no observed effect concentrations from chronic and subchronic tests with algae, daphnia and fish was divided by an arbitrary safety factor of 10 to derive the case study primary intended no effect concentration (PNEC). Potential risk for surface waters was then quantified by dividing the PECs by the PNEC. Potential risk from MPA was also assessed for sewage works and bacterial populations. In addition, MPA is not expected to bioaccumulate nor to adsorb to sewage sludge or to sediment to a significant extent. Conclusions on potential risks of MPA are given in the poster.

WE022 Cytostatics in Dutch surface water - overview of use and potential risks to the aquatic environment

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human medicinal products; at least 1200 of them are compounds of potential environmental concern. For the majority of these 1200 compounds data for an environmental risk assessment (ERA) are incomplete or lacking, with the result that their potential environmental impact cannot be assessed in an appropriate manner. The reason for this is simple: So called 'legacy products' have been authorised before the 'Guideline on the environmental risk assessment of medicinal products for human use' came into effect in 2006. According to the current legislation all 'legacy products' have to be reassessed for their environmental impact using an ERA. However, there are no statutory provisions in place to how to deal with legacy products. Hence, there is a vital need to prioritise active substances used in legacy products for further investigations and evaluation of their environmental impact as well as risk management activities. This is of particular importance because many of them are frequently detected in surface water and other environmental compartments. Moreover, active substances which are persistent, mobile, bioaccumulative and/or toxic have a specific mode of action as e.g. endocrine active substances, are in general of high environmental concern. We propose a step wise prioritisation concept that allows the identification of active substances with a high potential environmental impact and/or a high potential presence in the environment. The proposed tiered prioritization approach considers also elements of the EMA Guideline for environmental risk assessment of human pharmaceuticals. It is important to recognise that any approach needs to be flexible and adapt to the environmental changes and should be regularly adapted to the current state of knowledge.

WE025 SETAC Pharmaceuticals Interest Group
G. Maack, German Environment Agency / Ecotoxicological Assessment

WE026 What makes a chemical substance a 'natural substance'? A case study in the context of the EU veterinary medicines marketing authorisation procedure
T. Hahn, Fraunhofer Institute Toxicology and Experimental Medicine / Chemical Risk Assessment; C. Floeter, HAW Hamburg / Department of Environmental Engineering; S. Schnowbeck, G. Koennecker, Fraunhofer Institute Toxicology and Experimental Medicine / Chemical Risk Assessment. The marketing authorisation process for veterinary pharmaceuticals in the EU and other countries requires an environmental impact assessment (EIA) for each veterinary medicinal product (VMP). This EIA follows a phased approach with conservative estimates of environmental exposure in phase I, which may later require refinement by experimental data in phase II. Core of the phase I assessment is a catalogue of 19 questions on use and characteristics of the VMP under consultation. These questions aim at establishing an initial predicted environmental concentration, which, together with information on therapeutic use and targeted animal species, may trigger the necessity to perform an in-depth phase II assessment. According to question 2 in the phase I assessment a substance is exempted from further investigation when there is scientific proof that it is a natural substance “the use of which will not alter the concentration or distribution in the environment”. At first sight this definition appears unambiguous. Nevertheless may it be a hurdle for applicants because no further guidance is given which criteria apply for acceptance or rejection of a given concentration as 'natural'. Here, we present a case study comprising two substances in order to highlight possible uncertainties for applicant companies, as well as for competent authorities.

Obesogens and lipid disruptors (P)

WE027 Unraveling distinct pathways of PFOS toxicity by combining morphological, metabolomic and transcriptomic analyses
R. Longe, Institute of Environmental Assessment and Water Research (IDAEA-CSIC) / Environmental Toxicology; L. Navarro-Martín, C. Lucarelli, IDAEA-CSIC; E. Ortiz, IDAEA-CSIC / Department of Environmental Chemistry; A.E. Codina, CNAG, D. Raldua, IDAEA-CSIC; C. Barata, CSIC / Environmental Chemistry; B. Pina, IDAEA-CSIC / Department of Environmental Chemistry; R. Tauler, IDAEA-CSIC / Environmental Chemistry. Exposure to PFOS (perfluorooctyl sulfate) has been related to toxic effects on lipid metabolism, immunological response, and different endocrine systems. We present here a combined metabolomic and transcriptomic analysis of zebrafish embryos exposed to different concentrations of PFOS (30-1000 ppb) from 48h to 120 hfp. While parallel morphological analysis showed no macroscopic changes below the 1000 ppb mark, some metabolomic and transcriptomic changes occurred even at the lowest used concentration. Functional analysis of the observed changes revealed at least three major modes of action: alteration of PPAR signalling and lipid metabolism, effects on cell-cell interaction, perhaps linked to effects on the immune response and neuronal system development, and a general alteration of the development, reflected by an alteration of different development- and metabolism-related signalling pathways, likely affecting to cell cycle functions, and
to the metabolism of proteins, nucleotides, and amino acids. The results suggest a complex, multiple endocrine disruption-like toxic effects, at a concentrations well below the 1 ppm considered the LOAEC/NOAEC for many of the macroscopic effects traditionally linked to PFOS toxicity in zebrafish embryos, including lipid disruption, effects to sensorial organs, and lethality. It is also remarkable the functional correlation between these macroscopic effects and the molecular changes we observed at metabolic and/or transcriptomic levels at concentrations 10 to 100 below the macroscopic NOAEL.

WE028 Impacts of fatty acids and methylmercury on preadipocyte differentiation in rainbow trout (Oncorhynchus mykiss).

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Fish can be exposed to nutritional and chemical stress. In aquaculture, fish oil is increasingly replaced by plant-derived oils, which results in a modification of the fatty acid (FA) composition of the diet. Also, pollutants such as methylmercury (MeHg) are still present in aquatic environments. Adipose tissue is an essential endocrine organ involved in energy homeostasis and can be affected by some stressors. However, there is a lack of knowledge about the effects of FA and MeHg on rainbow trout adipose tissue. In this context, an in vitro experiment was conducted and linoleic acid (LA) induced a lipid content increase in fish, while MeHg decreased it. To understand better these results, two in vitro experiments were carried out on trout primary cultured adipocytes to study the effects of FA and those of MeHg. Effects of FAs - During 2 days, differentiation of confluent cells was induced through a hormonal cocktail. Cells were then incubated during 13 days with 0, 75, 150, 300 and 600 µM of α-linolenic (ALA), eicosapentaenoic, docosahexaenoic (DHA), LA, arachidonic and docosapentaenoic (DPA) acids and 2 mM MeHg in lipid mixtures. At day 13, for each FA, the higher the concentration, the more the lipid accumulation. At 600 mM, DHA and DPA were the most adipogenic FA, while LA and ALA (typical to plant-derived oils) induced less lipid accumulation. For all conditions, a clear enrichment of membranes and lipid droplets with the incubated FA was observed. Effects of MeHg - Confluent cells were incubated for 6 days with or without a hormonal cocktail, with 0, 0.5, 2.5 or 5 mM MeHg and with 4 µM lipid mixture. No cytotoxicity was observed. At day 6, cells were collected to determine mercury concentration, FA content and composition, and gene expression. Analyses are in progress. Preliminary results showed that the hormonal cocktail combined to increasing MeHg concentrations tended to affect cell morphology, towards a more typical adipocyte phenotype. In conclusion, it seems that the effects of FAs can be different at organism and cell levels. Regarding the effects of MeHg, we can highlight the presence of cells with typical adipocyte morphology in presence of hormonal cocktail and MeHg. For both experiments, analyses of expression of genes related to adipocyte differentiation, lipid metabolism and lipolysis are under progress and could provide helpful results to understand better the impacts of stressors in trout preadipocytes.

WE029 Obesogens in the aquatic environment

A. Capitão, CIIMAR; University of Porto; A. Lyssimachou, CIIMAR; E. Castro, CIIMAR - University of Porto; M.M. Santos, CIIMAR/FCUP / Biology/Endocrine Disrupters and Emerging Contaminants

The term of obesogen is a major health concern of our times, affecting an increasing proportion of the population worldwide. It is now evident that this phenomenon is not only associated with the lack of exercise and a balanced diet, but also due to environmental factors, such as exposure to environmental chemicals that interfere with lipid homeostasis. These chemicals, also known as obesogens, are present in a wide range of products of our daily life, such as cosmetics, paints, plastics, food cans and pesticide-treated food, among others. A growing body of evidences indicates that their action is not limited to mammals. Obesogens also end up in the aquatic environment, potentially affecting its ecosystems. In fact, reports show that some environmental chemicals are able to alter lipid homeostasis, impacting weight, lipid profile, signaling pathways and/or protein activity, of several lines of amphibians. Such a priority issue in aquatic toxicology. One of the most prevalent pharmaceutical contaminants is the type-2 diabetic drug metformin, which has been found in wide-ranging concentrations (ng/L - µg/L) in wastewater effluent and surface waters. Greater than 90% of metformin is metabolized into guanylurea during wastewater treatment, and it’s the metabolite guanylurea that is found in receiving waters in relatively high concentrations (µg/L). To improve our understanding of the toxicological effects of metformin and its metabolites, we demonstrated that about 1% of the waterborne concentration of metformin could be taken up in both embryonic and larval medaka after exposure windows ranging from 24 hours to 7 days. We also conducted a metabolism assessment of metformin and guanylurea exposed fish to elucidate the sub-lethal biochemical mode of action for each contaminant exposure. Significant changes were detected in the metabolome of 28-day larval male medaka exposed to both metformin and guanylurea, indicating significant dysregulation in fatty acid and lipid metabolism. This biochemical effect is likely a contributing factor to the observed decreased growth in exposed fish. In combination, these results suggest that the current concentrations of metformin and guanylurea in receiving waters are of ecotoxicological concern for resident fish populations.

WE030 Levels of proteins, carbohydrates, lipids and cholesterol in the digestive gland of juvenile catarina clam Argopecten ventricosus (Sowerby, 1842), exposed to toxic metals

A. Sobrino; Universidad Autonoma Metropolitana Iztapalapa; Universidad Autonoma Metropolitana; Universidad Autonoma de Baja California Sur

The analysis of the composition of the digestive gland, gives information on the energy level of the organism, this energy is mobilized in the different stages of its life cycle. When the organisms are subjected to severe stress conditions, it has been observed the mobilization of these reserves to maintain homeostasis, in short periods of time. In this work, an evaluation of the composition of the digestive gland of juvenile catarina clam exposed to the metals Cd, Cr, Pb and their mixtures was carried out to determine their energy content. Bioassays with water replacements were carried out. The organisms were exposed to 1 sublethal concentration of each metal (LC50) (0.35, 5.0 and 3.0 mg L−1 of Cd, Cr and Pb
respectively) and of the mixtures in proportion 1:1. The levels of proteins (Lowry, 1951), carbohydrates (Dubois, 1956), lipids (Bligh and Dyer, 1959) and cholesterol (Kit Biorad) were quantified at 24, 96, 144 and 168 hours after the start of the bioassay. The Kruskal-Wallis test showed that the difference between the concentrations of proteins, lipids, cholesterol and carbohydrates of the control group compared to the treatments was significant (p < 0.034). An increase in cholesterol levels was observed at 24 hours of exposure and a decrease in protein, lipid and carbohydrate levels of up to 75% in only 96 hours (4 days) of exposure to metals and their mixtures. This indicates that juveniles exposed to metals had high stress levels, (as was also observed in relation O:N). It should be mentioned that the surviving organisms of the tests, died 48 hours (2 days) after it was observed the mobilization of their energy reserves.

Environmental risk assessment and management of the spoil material produced in tunnelling excavation (P)

WE033 Environmental assessment of foaming agent persistence in conditioned soil for EPB-TBM tunnelling

The anionic surfactant sodium lauryl ether sulphate (SLES) is the main component of most commercial products used for soil conditioning in the excavation industry, in particular as lubricants for mechanized tunnelling. This excavation process produces a large amount of spoil materials that can have a potential impact on ecosystems. The lack of accurate information about SLES persistence in the environment has aroused increasing concern for their possible recycling as construction materials or as soil replacement for covering rocky areas. Currently, there are neither SLES soil threshold limits in European legislation, nor comprehensive studies on the environmental risk for soil ecosystems in these exposure scenarios. The objective of this study was to evaluate the biodegradability of the sodium lauryl ether sulphate (SLES) contained in two common commercial foaming agents (P1, P2). For this purpose, a set of microcosms was set up using two different soils (S1: silty-clay soil; S2: gravel in a clay-silty-sand matrix soil) sampled from a construction site. Moreover, the degradability of one product was evaluated in the presence of an additive used to improve the compactness and subsequently transport of the spoil material. For this purpose, microcosms were set up using soil samples conditioned separately with the two foaming agents. Control microcosms, consisting of untreated soil, were also set-up to compare the microbial community before and after the foaming agent addition. At selected times, soil samples were collected for assessing SLES concentration by ASE extraction followed by MBAS spectrophotometric method. Microbiological analyses were performed in order to assess microbial abundance, viability and dehydrogenase activity in the conditioned and control soils. Results showed that SLES degradation depended on the soil type, with DT50 ranging from 11 to 19 days; the additive increased significantly the surfactant persistence especially in the S2 soil. In the latter case, the higher persistence of the product can be ascribable to the detrimental effect of the additive on the microbial abundance and activity.

Application of the Vibrio fischeri acute toxicity test to assess the environmental impact of spoil materials containing foaming agents

The rapid development of TBMs in the tunnelling industry has been mainly due to their advantages over conventional excavation methods, such as continuous operation, safer working conditions, reduced damage at surface level and higher tunnelling speed. The performance of TBMs relies on the use of appropriate soil conditioning which consists in improving water solutions of anionic surfactants, mainly sodium lauryl ether sulphate (SLES) and in smaller concentrations other additives. In accordance with the Italian legislation, spoil material from excavation processes can be re-used as by-products if the chemical thresholds for organic and inorganic contaminants (e.g. heavy metals, hydrocarbons C>12; Italian Decree 120/2017) are not exceeded. However, there are currently neither SLES soil threshold limits nor Italian and Italian legislation (Annex 4 of the Italian Decree 120/2017), nor comprehensive studies on its ecotoxicological effects on soil and water organisms. The use of ecotoxicological tests makes it possible to overcome the analytical limits to detect multicomponent commercial foaming products (of which the complete composition is often unknown), to save time by avoiding the designing of new analytical methods for the increasing number of chemicals used in new foaming formulations continuously being put on the market. Above all, they provide information about the different interactions between the mixture and the specific matrix and the possible ecotoxicological effects on biota. In several studies aimed to evaluate the potential impact of spoil materials the bacterium Vibrio fischeri showed to be very sensitive to the residual concentrations of the surfactant SLES in eluates obtained from soil samples collected from excavation sites. The overall analysis of a set of chemical and ecotoxicological data showed that the bioaccumulation index was directly related to SLES concentration. Consequently, the ISO 11348-3:2007 test is a suitable tool to assess in a short time the occurrence of foaming agent residuals at effect concentrations in spoil material.

Biodegradability of the anionic surfactant sodium lauryl ether sulphate used as the main component in two foaming agents for tunnelling process
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New foaming agents with better environmental impact for tunnel excavation (P)
C. Testa, National Research Council of Italy / Water Research Institute; S. Raffo, National Research Council of Italy / Water Research Institute; M. Galeotti, National Research Council of Italy / Water Research Institute; A. Di Giulio, Italian National Research Council / Institute of Environmental Geology and Geoenengineering; P. Grenni, National Research Council of Italy (CNR) / Water Research Institute; T. Pescatore, National Research Council of Italy / Water Research Institute

Development of new foaming agents with better environmental impact for EPB soil conditioning - The Polyfoamer ECO line
A. Boscare, e. dal negro, Mapei SpA / Underground Technology Team; m. stefanoni, Mapei SpA / R&D

Biodegradability of the sodium lauryl ether sulphate (SLES) contained in two common commercial foaming agents (P1, P2) of the Polyfoamer ECO line. Various laboratory tests have been carried out with the new foaming agents of the Polyfoamer ECO line of products and samples of soil coming from different TBM projects. The results obtained with two samples of soil from an Italian project are described: the material called “M” (a

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Determination of anionic surfactants by Pressurized Liquid Extraction (PLE) followed by the modified Methylene Blue Active Substances (MBAS) method in spoil material from excavation processes


Recent years have been characterized by a rapid worldwide growth in underground constructions in the form of new infrastructures such as pipelines and communication cables as well as road and railway tunnels. The utilization of underground space offers a new strategy to urban planning, including a huge development of mechanized tunnelling industry by the use of Tunnel Boring Machines (TBM). Pressurized Liquid Extraction (PLE) is the most utilised method for the determination of appropriate soil conditioning products, principally foaming agents. Anionic surfactants are the main constituents of commercial foaming agents widely used as lubricating products in the mechanized excavation, improving the stability of the excavation face and reducing the friction between soil cuttings. Among the anionic surfactants, sodium lauryl ether sulphate (SLES) is the most utilised compound in the commercial products for the excavation industry. Significant amounts of rock broken into various sizes mixed with anionic surfactants are produced during the execution of engineering works. The potential re-use of this non-renewable natural resource, for example as land covering, depends on the assessment of its environmental compatibility; otherwise, the spoil materials must be treated as a waste. In this context, it is important to evaluate the residual concentrations of SLES in the excavated soils in order to evaluate their possible final destination. Given the absence of official analytical methods, it has become necessary to develop and validate a reliable and accurate methodology to quantify anionic surfactants in the spoil materials and, more in general, in the environmental solid matrices. For this purpose, the aim of the present work was the optimization of an analytical method for the determination of SLES in conditioned-soil samples. It consists of a first phase of extraction of anionic surfactants from the soil by the use of Pressurized Liquid Extraction (PLE) and the following analysis in the extract by the MBAS (Methylene Blue Active Substances) method using the water official method partially modified. The optimised method has been applied to real excavated soil samples because the determination of residual concentrations of the anionic surfactants in the spoil materials produced during excavation process, is currently one of the mandatory parameters for assessing their eco-compatibility.

Distribution and persistence of anionic surfactants in leachate and conditioned soil: mesocosm study for EPB-TBM applications


The development of the mechanized tunnelling industry by EPB-TBMs (Earth Pressure Balance - Tunnel Boring Machines), results in a wide use of foaming agents and polymers as lubricating products for soil conditioning. Anionic surfactants, and especially sodium lauryl ether sulphate (SLES) are the most used components of foaming commercial products. Soil debris from excavation processes can contain residual concentration of SLES. The potential re-use of the spoil material for public green areas or industrial purpose (e.g. land covering) depends on the site-specific SLES persistence in the excavated soil and on the related environmental exposure scenario. In this context, we evaluated the SLES leaching in different soils in order to simulate check whether underground water contamination may occur in a scenario where the spoil material is located close to a water body. For this purpose, we evaluated the persistence (DT₅₀) of SLES in two soils (S1: silty-clay soil; S2: gravel in a clay-sandy matrix soil) conditioned separately with two common commercial foaming agents, respectively F1 and F2, used at conditioning ratio giving final concentration in both the soils of about 150 mg/kg. The presence of strengthening foaming polymers (P1 or P2, 527 and 50 mg/kg respectively), needed in some cases to increase foam persistence, was also considered. After a preliminary phase at laboratory scale, a mesocosm experiment was conducted in order to entail the scale-effect, which is very significant when the soils are involved. SLES solutions were set-up mixing 100 kg of each soil with water, foam and polymer and then stored for 28 days in high-density polyethylene bins (HDPE diameter of 30 cm and height of 100 cm). The effect of soil type, grain size or aeration on SLES persistence was evaluated. For this purpose, eight bins containing S1 or S2 conditioned separately with the two foaming agents, were weekly turned to improve aeration, while the corresponding eight blends were not turned. At selected times (0, 4, 7, 12, 20 and 28 days), soil and leachate samples were collected from the bins for assessing SLES concentration by MBAS spectrophotometric method, preceded by ASE (Accelerated Solvent Extraction) in the case of the soil matrix. The results showed that residual SLES concentration in soil and in leachate is dependent both on the type of soil and on the nature of polymers.
agent products are anionic surfactants such as the alkyl ether sulphates (AES). The possible re-use of huge amounts of spoil material produced during the excavation process as by-products (e.g. land covering) or its discharge as a waste depends on the residual concentration of AES in the soil. The first option has the undoubted advantage to lower the costs of disposal. However, there are concerns about the potential environmental risk related to the re-use of conditioned soil. In fact, even if anionic surfactants are generally considered biodegradable and not toxic, there is little information in literature on their environmental fate and the possible ecotoxicological effects of the commercial formulations of foaming products and of the conditioned soils. The aim of this study was to evaluate the environmental compatibility and the ecotoxicological effects of two different soils treated with two different foaming agents containing the anionic surfactant AES, applying a suitable battery of bioassays. For this purpose, a set of soils has been prepared, containing two soils with different geopedological characteristics, conditioned with two foaming agents at the same treatment ratios (TR, L/m³) used for mechanized driller. Soil samples were collected at different maturation times (0, 7, 14, 28 days) in order to perform the ecotoxicological tests on the spoil material or in its aqueous extracts. The bioassays selected are representative of different trophic levels for the aquatic and terrestrial compartments: Microtox test with the bacterium Vibrio fischeri, Fish Embryo Acute Toxicity Test (FET) with the species Danio rerio, germination and growth test with the plant Lepidium sativum and test with the worm Eisenia fetida. In parallel, sub-samples of soil and eluate from each mesocosm were analyzed in order to determine the residual concentrations of the anionic surfactant AES. The overall results obtained showed different ecotoxicological response depending both on the kind of soil and on the PAE additive used in mechanized tunneling: effects on

WE041

Expedient test for on-site monitoring activity in mechanized tunneling applications


In the vast majority of tunnel projects performed with TBM-EPB technology a key issue is currently represented by the disposal management of the spoil, huge amount of excavated material mainly composed by the natural soil, water and chemical induces contaminants. The process to plan street disposal management in a virtuous cycle of reuse of the resources leads relevant economic and logistical advantages. The raising awareness about the chemical composition of the products injected during the excavation resulted in the development of experimental procedures aimed at studying the environmental impact of chemicals and their permanence in the soil during the excavation and after its completion. Commonly accepted standards have been developed to measure the level of pollution through expeditious tests directly on site. A joint research activity between Sapienza University and National Research Council of Rome has developed a test procedure able to provide expeditious information on the presence in the spoil of the chemicals often used in mechanized tunnelling. The results of preliminary laboratory tests convinced that the expedient assessment proposed can describe the amount of chemicals in the soil and their evolution in time, complementing the laboratory activities currently accepted. In fact, this fast procedure must be regarded as a first screening which can be run directly in site on a large number of samples without the use of expensive, delicate or complex instruments, to be used in combination with few more precise laboratory tests. Moreover, it seems to be particularly suitable for monitoring large volumes of waste involved in tunnel excavation. The intention for the future is to apply the procedure to real cases to verify, through a comparison with the most accurate laboratory tests, the actual effectiveness of this procedure.

WE042

Toxicity of some additives used in mechanized tunnelling: effects on daphnids, algae and cress.

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Surface agents and polymers are used in mechanized tunnelling to facilitate the excavation and to preserve the tunnel boring machine (TBM) from wear, block and break. As a by-product of the process, several tons of rocky debris are produced. These excavation rocks are made of rock fragments contaminated by the additives such as anionic surfactants, glycols, sealants, polyacrylates and polycrylamides. In the past, rocky debris were disposed as waste with a large economic impact on total excavation cost, while some disposal alternatives were recently applied, trying to recycle these complex materials as road filling material, covering rocky areas or river banks. These disposal solutions, however, have attracted the attention of regulators and environmental protection agencies, especially in Italy. In fact, the toxicity of these active mixtures is not yet fully known as well as the potential effectiveness in reducing from the simultaneous presence of additives with other regulated environmental contaminants such as, for example, metals and hydrocarbons. A preliminary study recently conducted by our group on three commercial TBM additives showed toxic effects on the aquatic ecosystem in concentrations comparable to those resulting from excavations carried out in Italy. This new study analyzes 8 surfactants and 4 commercial polymers, using a multidisciplinary approach to determine their reference thresholds for both water and soils, accounting of the effects on ecological targets. The chemical composition of the technical mixtures was determined by liquid chromatography coupled with high resolution mass spectrometry. The main chemical components were analyzed in silico to highlight the potential similarity with other pollutants, already listed in our environmental framework regulation. Finally, the toxicity of the various agents has been evaluated by tests with Daphnia magna, freshwater algae and cress. Chemical characterization identified 15 molecules present in all the surfactant mixtures, although in different proportion. No similarities with compounds already regulated by the Italian Environmental Act were found by the in silico analysis. All the tested surfactants were toxic for the aquatic organisms at concentrations comparable to those that can be found in leachates of conditioned rock debris. The additives resulted non toxic for the terrestrial plant at concentrations theoretically found in conditioned rock debris.


WE043

Bioaccumulation, tissue distribution, and trophic magnification of organic ultraviolet absorbent in freshwater ecosystem in the Pearl River catchment, China

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Bioaccumulation and trophic transfer of 13 organic ultraviolet absorbents (UVAs) were investigated in riverine wildlife organisms in the Pearl River catchment, South China. The UVAs were widely present in the fish with the highest level detected for UV 531. Generally, the UVAs concentrations were higher in the freshwater fish than in shrimp. Dietary habits of the fish showed effects on bioaccumulation of the UVAs with higher levels in the carnivorous species than in the herbivorous, planktivorous and detritus feeding fishes. Tissue distributions were generally in the order of liver > belly fat > muscle. Obviously higher concentrations of 2-ethyl-hexyl-4-trimethoxycinnamate were detected in eggs than in muscle, probably indicating maternal transfer of the compound. The calculated bioaccumulation factors (LogBA_FWere usually > 3.3, suggesting potential of the UVAs in the freshwater fish. UV 531 showed tendency of bioaccumulation in the fish from the sediment indicated by the calculated BSFA > 1. The estimated trophic magnification factors were > 1 for some benzotiazole stabilizers, suggesting potential of biomagnification of the UVAs in the freshwater fish.

WE044

Hyalella azteca as non-vertebrate alternative species for bioaccumulation studies

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Bioaccumulation is one of the PBT (persistence, bioaccumulation, toxicity) cut-off criteria for plant protection products (EC/1107/2009) in the EU; furthermore, high bioaccumulation is generally considered a critical parameter in other regions, too. The standard regulatory assessment of bioaccumulation is based on bioconcentration in aquatic species, i.e. for regulatory purposes in fish. However, standard fish bioconcentration studies are time consuming, expensive and they use a comparatively small number of compounds, while there is a need for a relatively quick, cheap, and preferably alternative test method that enables the ranking of structurally similar candidate molecules regarding bioaccumulation potential and the prediction whether a candidate molecule will exceed the BCF (bioconcentration factor) trigger value. Furthermore, Hyalella azteca might in the long-term perspective be able to replace fish for BCF testing. There is indication that experimental BCF values from flow through bioaccumulation studies with Hyalella are similar to those obtained from fish (Schlachtriem, 2012). Further work is presented in order to (i) increase the data base of Hyalella – fish BCF data sets covering a wide range of BCF values (i.e. 100 to 20 000), (ii) to standardize and simplify the test system and (iii) to check the suitability of the test system for molecules with an insecticidal mode of action which poses inherent challenges since Hyalella as an aquatic invertebrate can be quite sensitive. The results from
several BCF studies with _Hyalella_ supports this species as suitable test species for bioaccumulation testing and supports planned activities on OECD level. 

**WE045**

Bioaccumulation of ionizable organic chemicals in fish - The quest for reliable predictors

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Dietary bioaccumulation in fish is quantified using biomagnification factors (BMFs), which are derived under controlled conditions according to OECD guideline 305-III. To reduce in vivo experimental efforts, pre-screening using statistical models for BCF predictions is becoming increasingly popular. While dietary bioaccumulation of neutral chemicals has been successfully associated to lipoprophilicity descriptors, no suitable predictor has yet been identified for ionizable chemicals. In this study, we investigated the capability of selected chemical properties (e.g., molar volume, adsorption to albumin, lipophilicity, solubility, topological polar surface area) to predict bioaccumulation of organic electrolytes in fish with specific focus on dietary exposure studies. Measured dietary BMFs were collected from existing literature, and empirical correlations with measured or estimated chemical descriptors were evaluated. The dataset includes dietary BMFs in whole fish obtained under laboratory-scale conditions closely resembling or directly referring to the OECD 305-III guideline. In total, BMF data were available for 29 ionizable chemicals (of which 10 are perfluorinated chemicals), including 24 acids and 51 alcohols (to membrane lipids, structural proteins and albumin). A parallel dataset was compiled with bioconcentration factors (BCFs) of the same chemicals derived in water exposure studies with fish (OECD 305-1 guideline). Bivariative correlation analysis (Pearson and Spearman) revealed that a log_Kow was not a sufficient predictor of BMF, although with significant positive correlation (R^2=0.40), and that significant correlation was shown only with logK_pH of pH~1 to pH~3 (R<0.35). Furthermore, significant negative correlation was shown between BMF and solubility (R<0.60). These preliminary results indicate that commonly used predictors for bioaccumulation (e.g., log_Kow) are of limited relevance for ionizable chemicals, and other predictors should be identified. Ongoing research is focusing on the prediction of BCF from quantum-chemistry-based estimations of partitioning coefficients (tegmente/parameters lipophilicity and aluminum). Estimation of BCFs from BMF for the investigated chemicals will be also performed and verified with existing BCF measurements. Eventually, identified empirical regressions between BMF and chemical descriptors will be validated with ad hoc experimental data with radiolabelled test chemicals.

**WE046**

Evaluation of a tiered approach for the bioaccumulation assessment of fragrance substances: in silico, in vitro assays, invertebrate vs. in vivo fish bioconcentration test

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Bioaccumulation is a key end point in environmental hazard and risk assessment, especially for substances with a high octanol water partition coefficient (logKOW). To measure the BioConcentration Factor (BCF), a tiered approach is followed starting from the assessment of the octanol water partitioning coefficient as a measure for lipophilicity, which is often used as surrogate for lipid partitioning up to an experimental BCF value which is considered as the gold standard for fish bioaccumulation assessment. We have applied a series of non-animal approaches to predict the BCF values and compared those outcomes to the results from a BCF test in order to validate this alternative approach. Several fragments from the tetranorlabdane diterpenoids family, either composed of a single or a mixture of stereoisomers were tested. The logKOW predicted by QSAR ranges from 4.75-5.41 and at pH=3 and 2.2 when determined by HPLC (OECD 117). The slow stir method (OECD 123) provides a logKOW of 5.09 which is retained as the reference value. Various structure-activity relationship models were used to predict the fish bioconcentration factor, which ranged from ~ 1000 to ~ 4500, not exceeding the EU criteria for (very) Bioaccumulative substances (v>B), however, the structure was mostly outside the applicability domain of the models. Therefore in vitro assays were conducted on rainbow trout S9 fractions and hepatocytes confirming the potential of biotransformation; the refined BCF values calculated with IVIVE extrapolation models were <1000. In addition the bioaccumulation potential of one isomer was investigated in a flow-through test on the invertebrate _Hyalella azteca_ resulting in a BCFSS or kinetic < 500 L/kg. Finally an experimental fish BCF of ~500 (OECD 305) confirms that the fragrance composed of various isomers is not bioaccumulative, and supports the in vitro biotransformation findings. Histopathological results from toxicological studies showed liver hypertrophy consistent with the increased metabolism associated with detoxification processes. A tiered weight-of-evidence approach is clearly justified for the current bioaccumulation assessment, confirming that the tests described in the abstract may offer alternatives to animal testing when sufficient and supportive evidence is provided.

**WE047**

Proposal for a freshwater trophic magnification study based on a Comprehensive literature evaluation


Bioaccumulation potential is a critical property used for the risk assessment of chemicals and is usually expressed by parameters derived from laboratory or field experiments, in particular bioconcentration-, bioaccumulation- and biomagnification factors. A relatively new approach is the determination of so-called trophic magnification factors (TMF) which integrate enrichment processes in a food web to detect bioaccumulation in the food chain. The aim is to identify different types of biomagnification potential and to answer different questions in regulatory and monitoring affairs. The TMF can be used in the evaluation of the bioaccumulation potential of chemicals under REACH. However, TMF may be also applied in the context of the Water Framework Directive to normalize chemical monitoring data of fish to a common trophic level as well as to derive environmental quality standards for the protection goal ‘biovolume’. This study focuses on the comparison of a standardized experimental testing scheme (EAWAG 305) vs. a tiered approach that has already been performed and the investigated endpoints have shown considerable variation. The aim of this study is to define a sound concept for TMF investigations to enhance both, the reproducibility and accuracy of TMF estimates to allow the regulatory usage of this endpoint. The developed concept focuses on freshwater habitats, covers different invertebrate and fish species and will be tested in a pilot field study. A water body will be selected under consideration of several aspects such as the chemical burden of the water body, the type of contamination source, and species diversity in the water body. Invertebrate and fish species will be collected in the water body during spring/summer 2018. The trophic levels of the species will be determined applying different methods such as comparison of stable isotope patterns in the chemical and the non-chemical pathways (protein and lipoprotein). Estimation of BCFS from BMF for the investigated chemicals will be also performed and verified with existing bioaccumulation measurements. Eventually, identified empirical regressions between BMF and chemical descriptors will be validated with ad hoc experimental data with radiolabelled test chemicals.

**WE048**

Obstacles in identifying PBT/vPvB-substances under REACH for high tonnage chemicals

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The quest for reliable and valid biomagnification factors for underwater chemicals is an ongoing task. While the regulation of substances with PBT/vPvB properties are shortcomings in REACH, the proposal for a tiered approach for risk assessment and control of such substances is clearly justified for the current REACH regulation. A tiered approach which integrates results of alternative testing schemes and animal testing should be used to classify substances as PBT/vPvB or these substances are not covered by the REACH regulation (Regulation (EC) No 1907/2006. Registration, Evaluation, Authorisation and Restriction of Chemicals). The availability of that information in REACH registration dossiers of substances manufactured or imported in quantities of 100 to 1 000 tons per year (tpa) is evaluated in the current project on REACH compliance. This is a follow-up project on substances registered in quantities of 1 000 tpa or more. It is apparent that, the endocrine disrupting and developmental and reproductive toxicity of a substance is not considered and already the inorganic information on biotic and abiotic degradation, bioaccumulation and ecotoxicity are taken into account. The stepwise approach applied in the preceding project is adapted to the lower tonnage band with the aim to receive a high conclusion rate regarding the compliance of the endpoints. The previously published findings for substances registered in quantities of 1 000 tpa or more suggest that the main obstacle in identifying substances with PBT/vPvB properties are shortcomings in data quality, data gaps or inappropriate data-weighing/adaptation approaches. A minimum of 12% (for abiotic degradation) and a maximum of 61% (for ecotoxicity) of the dossiers were found to be “non-compliant”. It was recommended that registrants should thoroughly review and update their dossiers in order to fulfil the information requirements. This can be achieved either by using appropriate standard tests, providing a sound justification to waive data or using appropriate surrogate data. The poster will show preliminary results on the dossiers of substances registered in the tonnage levels of 100 to 1 000 tons per year and its
WE049 PBT/vPvB: All equally bad or some worse than others? - How to inform risk management
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In risk management, chemicals there is an increasing demand to assess the economic, social and environmental impacts of regulatory measures in order to facilitate political decision-making. Within REACH, socio-economic analysis (SEA) is the tool to assess and balance positive and negative impacts of different policy options. Currently, persistent, bioaccumulative and toxic (PBT) and very persistent, very bioaccumulative (vPvB) chemicals are regulated on the basis of their inherent properties. These imply a high and long-lasting damage potential for the natural environment. In reality, however, it is likely that PBT/vPvB are not of equal concern in terms of their damage potential to humans or ecosystems. The lack of knowledge on the actual effects of PBT/vPvB chemicals in the environment hampers the estimation of their risks and, in turn, a full quantification of all impacts. Consequently, a fundamental challenge for SEA is to adequately describe the regulatory concerns of PBT/vPvB chemicals, and to integrate specific information on a certain PBT/vPvB substance into a metric that informs policy-makers on their potential impacts. So far, there is no systematic approach on how available hazard and risk data as well as complementary information about the uncertainty due to data quality or lack of knowledge, can be used to assess the difference in damage potential of PBT/vPvB chemicals in SEA. Exploring the perspectives of experts from academia, industry and regulatory agencies may thereby help to guide the development of approaches for comparative evaluations of PBT/vPvB substances. This poster proposes a research project that is going to explore how specific characteristics of PBT/vPvB chemicals can influence the concern. Based on surveys and structured interviews, it will examined what parameters are considered most relevant to characterise the concern of PBTs/vPvBs. Furthermore, it will be explored how experts in academia, industry, regulatory agencies rate the relative importance of different (sets of) characteristics of PBT/vPvB substances in terms of their environmental impact potential. The results can be used to inform cost-effectiveness analyses and ranking schemes for PBTs/vPvBs. This will, ultimately, facilitate comparative evaluations of PBT/vPvB substances for SEA and regulatory decision-making.

WE050 Modelling Persistent & Mobile Organic Compounds using an updated Multimedia Urban Model: A Toronto Case Study with Organophosphate Esters (OPEs)
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Organophosphate Esters (OPEs) are a group of chemicals found at relatively high levels in the environment, in consumer products. The usage of OPEs has increased in recent years following the listing of penta- and octa-BDEs as POPs under the Stockholm Convention. In contrast with BDEs, OPEs highly polar molecules which can be considered persistent and mobile organic compounds (PMOCs). We modified the Multimedia Urban Model (MUM) of Diamond and co-workers by using polymolecular linear free energy relationships (ppLFERs) to represent partitioning, and it will be explored how experts in academia, industry, regulatory agencies rate the relative importance of different (sets of) characteristics of PBT/vPvB substances in terms of their environmental impact potential. The results can be used to inform cost-effectiveness analyses and ranking schemes for PBTs/vPvBs. This will, ultimately, facilitate comparative evaluations of PBT/vPvB substances for SEA and regulatory decision-making.

WE052 Polymers: The Next Frontier in Environmental Hazard Assessment
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Polymers are a very large and diverse class of chemicals widely used in cosmetic and personal care products. Their use and function are essential in creating high performing products that meet the needs of consumers. As used in cosmetic formulations, polymers can act as thickeners, emulsifiers, conditioners, opacifiers, film formers, rheology modifiers, etc. In the simplest terms, according to the Oxford Dictionary, a polymer is a substance that has a molecular structure consisting chiefly or entirely of a large number of similar units bonded together. They have a full range of physical-chemical properties including a wide breadth of solubility and molecular charge, for example. Currently, the majority of large molecular weight polymers are exempt from chemical regulations around the world (e.g. REACH) or are largely considered of low concern based on a minimum set of physical-chemical properties (US EPA's FIFRA, Australia's NICNAS). However, there is a speculation that these regulatory exemptions, specifically the REACH exemption, could be removed in the next 5-10 years. If this is the case, many previously untested chemicals would then need an environmental hazard assessment supported by an ecotoxicological dataset. This dataset may include aquatic toxicity testing, read-across to structurally similar chemicals that have been tested, weight of evidence toxicity estimates based on physical-chemical properties, or all of the above. However, the same variety of physical-chemical properties that allows polymers to have so many functions in cosmetic formulations also makes these substances difficult to test in aquatic systems – varying absorption properties, molecular charge, insolubility, etc. Therefore, safety assessors evaluating polymers must look to new and novel approaches for filling environmental data gaps in order to create a robust environmental hazard assessment. This poster will examine the current polymer landscape for cosmetic uses, identify common data gaps, provide possible solutions to fill those data gaps, and offer a prioritization scheme for future testing of polymers. Ultimately, the objective is to suggest a more modern approach to substantiating the environmental safety of the large variety of polymers used in cosmetic and personal care products.

WE053 A consistent Approach for PBT/vPvB Assessment for Pharmaceutical Products
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Keywords: Persistence, bioaccumulation, Toxicity, Pharmaceuticals Track 6: Environmental policy, risk management, and science communication. Session 6.7: PBT/vPvB Assessment: Update on regulatory guidance,
requirements, strategies and challenges. Abstract A PBT substance is one that is persistent (P), bioaccumulative (B) and toxic (T) or very persistent (vP) and very bioaccumulative (vB). The PBT assessment approach is well described under the REACH regulation (Regulation EC No 1907/2006) starting with a screening analysis based on available data and when a potential PBT is identified, then a definitive assessment is required. While this procedure is clearly understood for industrial chemicals, it is not regulated under REACh for pharmaceuticals. There is no clear definitive PBT/vP/B guidance for pharmaceuticals from the European Medicines Agency (EMA), although it is recommended that the assessment be made according to REACH criteria. Application of the REACH guidance to the PBT assessment of pharmaceuticals is not straightforward. A PBT evaluation of a substance is triggered within REACH if more than 10 tonnes of the substance is used per year. There is no established trigger value for performing a PBT assessment for pharmaceutical products, although it appears that a PBT assessment is applicable to pharmaceutical products that go into Phase II. However, our experience is that there is room for interpretations during the review process – especially for products that end at Phase I. Some RMS have consistently rejected the use of all available data; especially data derived from QSARs and instead treat the product as non PBT. Furthermore, the information on the regulatory consequences of the PBT-assessment for any given product, the situation may change in the future. It is our hope the EMA will issue clear guidance on how a PBT/vP/B assessment should be performed for pharmaceutical products and the consequences for products which fulfill the PBT/vP/B criteria. This presentation will describe our experiences and the challenges we have encountered in performing PBT/vP/B assessments for pharmaceutical products, including an overview of typical review comments from Co-(Rapporteur) are discussed. We also discuss review options for the improvement of PBT/vP/B assessment for pharmaceuticals.

WE054 Evaluation of new assessment methods and enhancement of PBT/vP/B criteria for ionisable substances

H. Holzmann, RWTH Aachen University; A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics

The identification of persistent (P) bioaccumulative (B) and toxic (T) substances under the EU regulation REACH (Registration, Evaluation, Authorisation and Restriction of Chemicals) was developed to assess neutral organic compounds. However, nearly 50 % of the chemicals pre-registered at the European Chemicals Agency (ECHA) are partly or completely ionised under environmental conditions (Franco et al., 2010). Since the charge of chemicals strongly influences their properties and environmental behaviour, the currently valid concept under the REACh regulation does not provide a sufficient basis for the assessment of ionisable substances. The objective of the project is to refine the P assessment of ionic and ionisable substances under REACH. For this purpose, simulation tests following OECD guidelines are conducted using two different types of environmental compartments: Aerobic and Anaerobic Transformation in Aquatic Sediment Systems (OECD 308) and Aerobic Mineralisation in Surface Water (OECD 309). As models we have encountered in performing PBT/vP/B assessments for pharmaceutical products, including an overview of typical review comments from Co-(Rapporteur) are discussed. We also discuss review options for the improvement of PBT/vP/B assessment for pharmaceuticals.

WE055 Assessment of the persistence of ionisable or ionic organic chemicals under REACH

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Persistence of humans and the environment, the identification and regulation of chemicals with persistent (P), bioaccumulative (B) and toxic (T) properties are central within the environmental assessment. The criteria for the identification of PBT substances under REACH (Registration, Evaluation and Authorization of Chemicals) (EU Nr. 1907/2006) and the guidance for the PBT-assessment have been developed for neutral organic molecules, and do not properly address charged chemicals. Due to their charge, ionic and ionisable substances seem to behave differently in the environment compared to neutral substances. With the addition of cationic, anionic or amphoteric characteristics, the chemicals intrinsic properties (e.g. water solubility, log Kow) change as a function of the environmental pH. This dependency affects the distribution of these substances within environmental compartments. The ionic function may also lead to different interactions between organic or mineral solid particles and the substance, influencing their bioavailability for potentially decomposing microorganisms, which are governing biological degradation. In order to improve the evaluation of the persistence of ionic and ionisable substances in the PBT-assessment, sorption and degradation patterns of the ionic or non-ionic substance carrying either a positive, negative or non-charged functional group will be investigated. An example of non-charged behaviour of 14C-labelled 4-N-Dodecylenephelenol, 4-n-Dodecylbenzenesulfonicacid sodium salt and 4-8-Dodecylbenzyltrimethylammonium chloride in soil simulation tests (OECD 307) will be investigated according to their mineralized, extractable and non-extractable fraction. Non-extractable residues will be investigated further. In addition, the sorption behavior of the model substances will be determined in a sorption study (OECD 106). The results will enable the estimation of the effect of the positive and negative charged groups within the molecule structure regarding the biodegradability and will improve the evaluation of the persistence of ionic and ionisable substances in the PBT-assessment.

WE056 Interaction of sulfonamide with soil humic acid: ESIs investigations with nitroxide spin label

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Surface water, about 40 % (DS-1) of the pharmaceutical products, including an overview of typical review comments from Co-(Rapporteur) are discussed. We also discuss review options for the improvement of PBT/vP/B assessment for pharmaceuticals.

WE057 The role of non-extractable residues in the environmental risk assessment from regulatory perspective - requirements and challenges

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In previous studies, we observed different results in different regulatory frameworks. In some cases NER are integrated in the calculation of predicted environmental concentrations (PEC) or are an issue in the authorisation decision. The significance of NER in the assessment of persistence (e.g. PBT, vB/P, POP classification) has been more or less neglected in the past. However, new developments as reflected in guideline revisions (e.g. ECHA R.11, 2017) highlight the importance of NER in toxicity assessment and sorption. NER can be reversibly bound to the soil/sediment and pose a potential risk to the environment or irreversibly bound which can be interpreted as sink. Hence, the potential release of parent or transformation products from NER in soil or sediment should be considered. However, distinguishing between these types of NER presents a challenge up to now. Standardised or commonly accepted extraction schemes or analysis techniques are not available due to the broad range of substances and soil/sediment characteristics. A general classification for NER was proposed by Eschenbach (2013) based on a literature survey dividing NER into four types: type 1 (heavily sorbed fraction) and type 2 (physically entrapped fraction) are
considered to be possibly remobilised in the environment. NER Type 3 (covalently bound residues) and type 4 (biogenic bound NER) are considered to be irreversibly bound to soil/sediment or transformed into biomass and therefore a risk to the environment is not anticipated. Furthermore, a stepwise extraction scheme is proposed which would allow the determination of these different NER types. The comprehensive scientific assessment of this extraction scheme supported by experimental research was conducted by UBA.

Transformation tests in soil with 13C-labelled substances were carried out in accordance with the OECD 307 guideline. Different extraction methods and chemical breakdown procedures were performed and compared in order to characterise the formation of the different NER types. Under consideration of these results, a refined extraction scheme will be proposed with respect to the general applicability for the formation of NER. This will be used for an assessment of reversibly bound fractions have to be considered in assessment of the persistence.

An UBA evaluation of regulatory data demonstrate the effects of several influencing factors (e.g. extraction methods, soil type) on NER formation.

**WE058**

**Sorption properties of Ionic organic chemicals: Correlations between ion exchange chromatography retention factors and environmental sorption coefficients**

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A common metric for the extent of equilibrium sorption and partitioning, such as the octanol-water partition coefficient (Kow) for neutral organic chemicals, does not exist for ionic organic chemicals. Finding a suitable reference sorption system for ionic organic chemicals should enhance the evaluation, modeling, and prediction of environmentally-relevant sorption coefficients for such chemicals. In this study, we considered synthetic ion exchange materials as possible reference systems to characterize the formation of the different NER types. Under consideration of these results, a refined extraction scheme will be proposed with respect to the general applicability for the formation of NER. This will be used for an assessment of reversibly bound fractions have to be considered in assessment of the persistence.

**WE059**

**Simulation of the fate of co-labeled 13C3-15N-glyphosate in a water-sediment system and formation of biogenic non-extractable residues**

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The combination of dynamic simulation and stable isotope techniques allows tracking the assimilation of pesticides into biomass [1]. Here, we simulated the fate of co-labeled 13C3-glyphosate in an Outdoor Water degradation test [2].

The mathematical model used consisted of two compartments for sediment (slow and rapid ad-desorption), one compartment for dissolved mass, and microbial growth and metabolism. The flow of both 15N and 13C were balanced. The model considers two biodegradation pathways for glyphosate, namely the sarcosine-pathway with complete mineralization, and the incomplete pathway with ANA, non-stable bound or homolysis of the ring. The microbiological production was partly estimated from the data, while others were calculated. The microbial growth yield was predicted from the MTB method, using thermodynamics and chemical structure [3]. The model can capture the dynamics of the system, including degradation of glyphosate, formation of AMPA and CO₂, formation of living and dead biomass (proteins) and chemical adsorption. At the end of the experiment (80 days), non-extractable residues accounted for 23% of the 13C and 26% of the 15N. 10% of the 13C and 12% of the 15N were recovered from the protein fraction (mostly non-living amino acids), which is equal to the biogenic non-extractable residues (NER). Biogenic NER consisted of assimilated 13C/15N and are thus considered to be irreversibly bound as proposed in the updated ECHA guideline for PBt/PvP assessment [4]. This is the first study simulating the formation of biogenic NER using experiments with 15N-labeled molecules. [1] Kästner, M., Nowak, K. M., Miltner, A., Trapp, S., & Schiffer, A. (2014). Classification and Modelling of Nonextractable Residue (NER) Formation of Xenobiotics in Soil – A Synthesis. Crit Rev Environ Sci Technol, 44(19), 2107–2171. [2] Wang, S., Seiwert, B., Kästner, M., Miltner, A., Schiffer, A., Reimersma, T., Q. Yang, Nowak, K. M. (2016). (Bio)degradation of glyphosate, amino acids and a stable isotope co-labeling approach. Water Res., 99, 91–100. [3] Brock, A. L., Kästner, M., Trapp, S. (2017). Microbial growth yield estimates from thermodynamics and its importance for degradation of pesticides and formation of biogenic non-extractable residues. SAR QSAR Environ Res, 28(8), 629–650. [4] European Chemical Agency (2017). Guidance on Information Requirements and Chemical Safety Assessment. Chapter R.11: PBt/PvP assessment, Helsinki, Finland.

**WE060**

**Determination of persistent organic pollutants (POPs) in soil from sites adjacent to landfills: different provinces of the Republic of Armenia**

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Sources of environmental pollution by persistent organic pollutants (POPs), either used or stored, ceaselessly applied as pesticides include landfills, many of which do not meet sanitary and hygienic standards and, at open burning of wastes, become a source of dioxins and furans. Investigation was carried out on soil samples taken in different marzes (provinces) of the Republic of Armenia at the boundaries of landfills and agricultural lands or water basins in the vicinity of towns Ararat (Ararat Marz), Hrazdan (Kotayk Marz), Sevan (Gegharkunik Marz), Gavar (Gegharkunik Marz), Aragatz (Ararat Marz). The obtained soil samples were analyzed for determination of the following POPs: - Hexachlorocyclohexane (HCH) isomers: α-, β-, γ-; - HCH: 2,4'-DDT: 2,4',4'-DDT; - DDT metabolites: 4,4'-DDE, 4,4'-DDD, 2,4'-DDD, 4,4'-DDE: - Hexachlorobenzene, - Heptachlor, - Aldrin - Dieldrin - Heptachlor epoxide A and Heptachlor epoxide B - Endosulfan I and Endosulfan II - Endrin, - Dinitro- Dioxin-like polychlorinated biphenyls: congeners No. 77, 81, 105, 114, 123, 126, 156, 157, 167, 169, 180, 189. Quantification of POPs was done using chromatograph with electron capture detector (ECD) equipped with glass capillary column with stable phase DB-5MS UI and the following parameters: 60 m x 0.25 mm x 0.25 μm. Special attention was paid to the total concentrations of HCH isomers, DDT isomers and their metabolites, as well as the total amount of polychlorinated biphenyls, as maximum allowable concentrations (MACs) are set for the aggregate amount of these compounds. On the analogy, concentrations of other POPs pesticides and all studied POPs are also considered on the whole (summarization concentrations) as obvious integral indicators of soil pollution by the studied POPs. In the investigated soil samples HCH isomers, DDT isomers and metabolites, certain Dioxin-like polychlorinated PCBs were detected at concentrations exceeding the established norms.

**WE061**

**Improving the interpretation of Non-Extractable Residues (NER) in degradation assessment**

A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics; M. Kaestner, Helmholtz Centre for Environmental Research UFZ / Dept Environmental Biotechnology; S. Trapp, Technical University of Denmark DTU / DTU Environment

Criteria for the assessment of chemical properties, potential (eco)toxicity, and environmental behaviour of industrial chemicals in general, and particularly for REACH substances, biocides, pesticides, and veterinary medicines are summarized in specific European legislations. The regulatory views on NER formation differ considerably, with two extremes of assuming them as either degraded residues of environmental behaviour of industrial chemicals in general, and particularly for REACH substances, biocides, pesticides, and veterinary medicines are summarized. Type II NER are residues that are covalently bound to organ for type II this will rarely happen under physiological conditions. Type III NER comprises biogenic NER (bioNER) after complete metabolization of the xenobiotic for type II this will rarely happen under physiological conditions. Type III NER comprises biogenic NER (bioNER) after complete metabolization of the xenobiotic and the incomplete pathway with complete mineralization, and the incomplete pathway with ANA, non-stable bound or homolysis of the ring. The microbiological production was partly estimated from the data, while others were calculated. The microbial growth yield was predicted from the MTB method, using thermodynamics and chemical structure [3]. The model can capture the dynamics of the system, including degradation of glyphosate, formation of AMPA and CO₂, formation of living and dead biomass (proteins) and chemical adsorption. At the end of the experiment (80 days), non-extractable residues accounted for 23% of the 13C and 26% of the 15N. 10% of the 13C and 12% of the 15N were recovered from the protein fraction (mostly non-living amino acids), which is equal to the biogenic non-extractable residues (NER). Biogenic NER consisted of assimilated 13C/15N and are thus considered to be
II NER and type I NER should be considered as potentially releasable residues in persistence assessment but the probability of type II release is much lower. For these types the potential of remobilization needs to be tested and evaluated. Our concept is to consider the total amount of NER minus bioNER as the amount of potentially remobilisable xenonNER (type I + II). If a clear differentiation of type I and type II is possible and the latter is irreversibly bound, only type I NER needs to be considered in the persistence assessment. If no characterization of NER is available, we recommend to assess the total amount as potentially remobilizable.

The use of pesticides has allowed for increased crop productivity in agriculture. However, because of their frequent application onto agricultural lands, their environmental fate is of particular interest. Pesticides have been ubiquitously detected in natural waters throughout Europe as well as in water supplies, which suggests these pesticides may be more mobile and persistent than their initial evaluations indicated. A potential reason for the mischaracterization of these pesticides' persistence could be the abiotic degradation pathways in the environment. For example, only direct photolytic in pure water can be considered, but the presence of dissolved organic matter can significantly affect the photolytic fate of pesticides in natural water. The photolytic degradation can either be enhanced by indirect photolysis reactions with photochemically produced reactive intermediates (e.g., triplet state dissolved organic matter, single oxygen, hydroxyl radical), or it can be suppressed by the light screening effect of organic matter once the light enters the water column. This research focused on the photolytic fate of fungicides, which account for approximately half of pesticide usage in Europe, in natural waters. For the several commonly used fungicides investigated, the results show that both direct and indirect photolysis are important to quantify in order to accurately characterize the environmental fate of fungicides.

Substances of unknown or variable composition, complex reaction products or biota. In this case UV absorbs data is provided for the extrapolation which cannot be fully identifiable and their composition may be difficult to predict. Under REACH Registration, it may be necessary to test these substances, which from many perspectives represents significant challenges. Reference is made to REACH Regulations and in particular Annex VII Section 9.2 where a series of degradation studies are proposed including simulation testing in surface water, soil, and sediment. The study should be with an endpoint in each case including identification of degradation products. It is relatively straightforward to generate e-fate data on a substance of known composition (chemical identity and purity) based on results from simulation studies in soil, sediment or water. OECD test guidelines 307, 308 and 309 used to describe experimental designs for simulation testing require the use of high purity material (>95%) and the use of a radiolabelled substance is highly recommended if the researcher aims to study the degradation products. It is therefore fundamentally not possible to perform simulation tests to determine the e-fate characteristics of UVCB substances due to the variable, unknown or unpredictable nature of the starting material. If testing of the "substance" is flawed then there are 2 options - the first is to isolate, purify and identify individual substances from the UVCB and then determine the e-fate characteristics of each component independently of each other...this seems unrealistic in most cases. Incorporation of a radiolabel to this test is likely to be prohibitive in terms of scale. An alternative approach would be to consider the chemical structures in a UVCB in groups and to test exemplar molecules or structures from within the mixture. This approach is possible when the individual structures have similar physico-chemical properties and structures are related. In this case UV adsorbance data is provided for the exemplar which can be used to predict the overall persistence of the UVCB substance. Choosing the most appropriate exemplar molecule may be challenging and examples are given. If the exemplar molecule is persistent then reasoned logic would dictate that the UVCB substance was persistent and further testing implemented accordingly.

The thermodynamic photodegradation half-lives of atrazine at depths in natural waters at different depths were calculated based on its molar absorption spectrum, quantum yield, and the type of exchangeable cation present on montmorillonite. Based on our results, the decay of atrazine can be used as an example, and used laboratory study results to describe experimental designs for simulation testing require the use of high purity material (>95%) and the use of a radiolabelled substance is highly recommended if the researcher aims to study the degradation products. It is therefore fundamentally not possible to perform simulation tests to determine the e-fate characteristics of UVCB substances due to the variable, unknown or unpredictable nature of the starting material. If testing of the "substance" is flawed then there are 2 options - the first is to isolate, purify and identify individual substances from the UVCB and then determine the e-fate characteristics of each component independently of each other...this seems unrealistic in most cases. Incorporation of a radiolabel to this test is likely to be prohibitive in terms of scale. An alternative approach would be to consider the chemical structures in a UVCB in groups and to test exemplar molecules or structures from within the mixture. This approach is possible when the individual structures have similar physico-chemical properties and structures are related. In this case UV absorbance data is provided for the exemplar which can be used to predict the overall persistence of the UVCB substance. Choosing the most appropriate exemplar molecule may be challenging and examples are given. If the exemplar molecule is persistent then reasoned logic would dictate that the UVCB substance was persistent and further testing implemented accordingly.

Fate Testing of UVCB Substances

E. Papa, University of Insurbia / Department of Theoretical and Applied Sciences (DiSTA); L. Motto, University of Insurbia / Department of Theoretical and Applied Sciences DiSTA; A. Sangion, P. Gramatica, University of Insubria / Department of Theoretical and Applied Sciences (DiSTA); M. Minella, D. Vione, University of Torino / Chemistry

Chemical reactions driven by sunlight are important processes in surface freshwaters, where they are involved in the transformation of xenobiotic molecules and of naturally occurring compounds. The relevant reactions are generally divided into direct photolysis, which involves molecules that absorb sunlight and are transformed as a consequence, and indirect photolysis, which involves reactive transients such as OH, CO₂, O₃, and the triplet states of chromophoric dissolved organic matter (CDOM*). They are generated by irradiation of photosensitisers such as CDOM (producing CDOM*, O₃, and OH), nitrate and nitrite (producing OH). Among these transient species, CDOM* is certainly the most reactive one in terms of its number (which is a consequence of the poorly known nature of CDOM) and reactivity. Still, CDOM* is involved into the transformation of several organic pollutants. In this work different triplet sensitizers that may be used as surrogates to estimate second-order rate constants with CDOM have been studied in silico. In particular, the experimental second-order reaction rate constants measured for the photodegradation of 49 pesticides commonly used as CDOM proxies (1-nitronaphthalene (1-NN), riboflavin (Rb), 4-carboxybenzophenone (4CBP), and anthraquinone-2-sulfonate (AQS)) have been used to derive Quantitative Structure-Activity Relationships on the basis of theoretical molecular
descriptors. The choice of phenols is motivated by the fact that they are the most likely compounds to undergo triplet-sensitised phototransformation in sunlight surface waters. Results show that the reaction rate constants with 3AQ2S and 4ICBP give the best QSAR models that can be used to simulate the photodegradation of phenols and similar compounds in the presence of CDOM. Quality differences in the QSARs generated for these reactions are probably due to differences in the chemical structure of the four sensitisers. These results provide additional insight into the mechanisms that regulate the fate of potential organic pollutants in surface waters. They will be used to design future experimental tests by focussing on one/two among the studied sensitisers, and to predict the photodegradation of new and existing substituted phenols and similar compounds on the basis of their chemical structure.

WE067
In silico Tools to Assess the Confidence of QSAR Model Predictions
R. Kühne, Helmholtz centre for environmental research - UFZ / Department of Ecological Chemistry; S.S. Kutsarova, O. Mekenyun, University of Zlataros / Laboratory of Mathematical Chemistry; G. Schuurmann, Helmholtz centre for environmental research - UFZ / Department of Ecological Chemicals, Helmholtz Centre for Environmental Research; J. Raether, Laboratory of Mathematical Chemistry; G. Schuurmann, Helmholtz Centre for Environmental Research – UFZ / Laboratory of Mathematical Chemistry.

For the regulatory acceptability of QSAR predictions solid information about the reliability of the applied models is crucial. This regards the model in general as well as the particular prediction for a certain chemical. The presented study provides computerized tools to support the assignment of prediction reliabilities. The first topic of concern is the applicability domain. Generally, the applicability domain comprises various aspects. The focus of this presentation is on the mechanistic domain and on the chemical domain in terms of structures and substructures. In particular, the application of atom-centered fragments (ACF) is demonstrated. While ACF characterization of the general structural domain of a training set is established already, the approaches shown here provide more specific information about the reliability of a prediction. On one hand, this is achieved by extending the ACF fragment based sets beyond the training set. Furthermore, data sets are separated into subsets with regard to performance or value ranges, and these subsets are employed to derive advanced reliability indicators. Secondly, automatically obtained model selectors can assist in selecting the presumably best-performing model from a model suite for a certain chemical depending on its structure and properties. Model selectors in this regard are computed scores derived from application of the model suite to chemicals with known experimental data. Thirdly, consensus modelling strategies are presented and examined to compensate for individual model errors. When combining predictions from different models, consensus outcomes can accordingly increase the levels of confidence, while conflicting outcomes are indicating lower reliabilities. In this respect, specific indices are being adapted for the in silico method of Ecological Chemicals suites, and consensus approaches contribute to weight of evidence assessments. For all three aspects, working tools will be presented, and their performance will be demonstrated via examples from existing models and data sets. Acknowledgment: This study was financially supported by the European Union 7th Framework Program SOLUTIONS (FP7-ENV-2013) of the under grant agreement no. 603437.

WE068
Data Gap filling with ECOSAR in K-REACH compliance, a limitation and weakness
J. Raether, Korea Institute of Industrial Technology / Environmental Science and Engineering; H. Park, Korea Institute of Industrial Technology; S. ok, Kitech / Research Center for Environment & Risk assessment.

ECOSAR is a computer based QSAR program developed by US EPA within the regulatory constraints of the TSCA. However, it is also used in the other country or organization such as EU, Korea, OECD, etc. for their regulatory purpose. We introduced ECOSAR program to generate toxicity data and fill the data gap for developing species sensitivity distribution of 20 organic compounds. However, ECOSAR shows more pragmatic than theoretical characteristic. Thus, we investigated whether this model shows acceptable results on the deficient data of 20 organic compounds or not. Therefore we collected published data for fish and daphnia and compared their geometric mean value. On the contrary, methyl hydrazine shows almost but others s...
article 57(f) of REACH based on a demonstrated equivalent level of concern (ELoC) as PBT or vPvB substances. In order to identify PMT as a substance of ELoC it must be demonstrated that there is “scientific evidence of probable serious effects to human health or the environment which give rise to an equivalent level of concern” and that there is evidence from a risk based considerations that the substance may cause serious effects during use and after through consideration, it should be expected that the substance has a high potential for the candidate list is the most effective strategy management strategy. With the protection of drinking water and pristine water bodies in mind, both bioaccumulation and mobility of persistent chemical substances are non-desirable. The environmental effect felt by bioaccumulative and mobile chemical substances varies both temporally and spatially, where mobile compounds can potentially accumulate in semi-closed drinking water cycles, while bioaccumulative compounds are able to accumulate in a closed loop system. vPvB can accumulate in food chains over time while vPvM can accumulate in pristine environments over time.

**WE072**

How many vPvM/PMT substances have been registered under REACH? - vPvM/PMT screening by using the Danish QSAR database

R. Holberg, Danish EPA / Chemicals; E.B. Wedesby, N.G. Nikolov, Technical University of Denmark (DTU) / Division of Diet, Disease Prevention and Toxicology, DTU Food; K. Tyle, DK EPA / Chemicals

UBA, Germany, has initiated work to develop criteria to identify substances which are very persistent and very mobile (vPvM), and persistent, mobile and toxic (PvM) and substances with a potential for contamination of ground water with mobile (M) and very mobile (vM) substances. Two screening algorithms were developed using the substance properties of water solubility (Sw) and the soil/sediment organic carbon-water partition coefficient (log Koc) by equating log Koc and Sw values based on >64,000 substances. The pH-dependent octanol-water partition coefficient (log Dow) was used as an alternative value for substances which ionize at environmentally relevant pHs. For ecotoxicity, similar QSAR algorithms as used for the ecotoxicology screening under PBT assessments were used. In addition, new algorithms for predicting CMR properties from ongoing work on the updated Advisory Self-classification List (expected to be published by end of 2017 by the Danish EPA) were included to also take potential chronic mammalian toxicity into account. The selected QSAR based T-related algorithms were employed on top the screening algorithms for P and M properties as a screening tool for substances with a potential for mammalian and non-mammalian toxicity to wildlife species and human health. The results of the screenings identify the potentially vPvM and PBT substances currently registered under REACH in tonnages > 10 tpa per manufacturer or importer. The screenings identify substances according to criteria proposed by UBA and the developed QSAR algorithms were also evaluated on top the screening algorithms for P and M properties. The results provide input for current and future work with the concept of vPvMs/PMTs. The Danish QSAR DB contains 650,543 substances, of which 80,085 currently are pre-registered and/or registered under REACH. Future vPvM/PMT screenings can be refined to address specific substance groups of interest; substances registered after the last REACH registration deadline; or address future modifications, if relevant, in the proposed vPvM/PMT criteria.

**WE073**

Identifying PMT substances amongst REACH registered substances

H. Ary, NGI / Environmental Technology; S. Hale, Norwegian Geotechnical Institute; A. Striffler, denkbares; D. Sätterl, UBA / Section IV; Chemicals; L. Schliebner, UBA; M. Neumann, German Environment Agency (UBA) / Section IV 2.3 Chemicals

The chemicals that have the greatest chances of appearing in drinking water are those that are mobile in the aquatic environment enough to enter drinking water sources and persistent enough to survive water treatment processes. Despite the growing number of organic compounds released into the market, there is very little consideration as to how to identify or categorize which of them are persistent, mobile and toxic, (PMT) and thereby pose a potential threat to drinking water. The list of REACH registered substances as of May 2017 was independently evaluated for their likelihood of being a PMT. The evaluation of persistent (P), very persistent (vP) or potentially persistent (i.e. Pcscreen) was performed according to REACH guidelines. For mobility, a criteria of a measured (or estimated) log Koc of six or less fluorinated carbons (“short chain”). Fluorotelomer-based products can be in either the polymeric or non-polymeric PFAS categories. Within the polymeric PFAS category, the fluorinated repellent products, including durable water repellents (DWRs), are found. These are normally side-chain fluorinated polymers typically applied in combination with other finishing auxiliaries. The side-chain polymeric fluorotelomer-based products perform exceptionally well and provide unique chemical and critical properties on high-end performance garments, workwear, first responder gear and non-water based fluorochemical PFAS category, fluorotelomer-based surface active agents (e.g. “fluorosurfacants”) are used in complex multi-component formulations such as Cleaning Products, Paints, Coatings and Aqueous Film Forming Foams (AFF). The non-polymeric fluorotelomer-based products provide superior surface wetting and leveling properties and are very mobile. Due to their high concentrations in surface and ground water drinking as drinking water (human health concern). QSAR screenings using the free online Danish QSAR DB were performed on 2,372 mono-constituent organic substances. For persistency (P) algorithms as used for the persistency screening under PBT assessment was used. A new screening algorithm for very persistent substances (vP) was set up by adjusting the P screening algorithm. For mobile (M) and very mobile (vM) screening algorithms were developed using the substance properties of water solubility (Sw) and the pH/electrolyte organic carbon-water partition coefficient (log Koc) by equating log Koc and Sw values based on >64,000 substances. The pH-dependent octanol-water partition coefficient (log Dow) was used as an alternative value for substances which ionize at environmentally relevant pHs. For ecotoxicity, similar QSAR algorithms as used for the ecotoxicity screening under PBT assessments were used. In addition, new algorithms for predicting CMR properties from ongoing work on the updated Advisory Self-classification List (expected to be published by end of 2017 by the Danish EPA) were included to also take potential chronic mammalian toxicity into account. The selected QSAR based T-related algorithms were employed on top the screening algorithms for P and M properties as a screening tool for substances with a potential for mammalian and non-mammalian toxicity to wildlife species and human health. The results of the screenings identify the potentially vPvM and PBT substances currently registered under REACH in tonnages > 10 tpa per manufacturer or importer. The screenings identify substances according to criteria proposed by UBA and the developed QSAR algorithms were also evaluated on top the screening algorithms for P and M properties. The results provide input for current and future work with the concept of vPvMs/PMTs. The Danish QSAR DB contains 650,543 substances, of which 80,085 currently are pre-registered and/or registered under REACH. Future vPvM/PMT screenings can be refined to address specific substance groups of interest; substances registered after the last REACH registration deadline; or address future modifications, if relevant, in the proposed vPvM/PMT criteria.

**WE074**

Recent Advances in Toxicology, Safer-Alternatives Assessment, Value-in-Use and Best Practice Guidance of Short-Chain Fluorotelomer-based Products for AFFF, Textiles and Other End-Uses

S. Korzeniowski, BeachEdge Consulting; J. Bowman, FluoroCouncil Per- and polyfluoralkyl substances (PFAS) is a term that describes a wide and diverse array of chemistry containing fluorine and carbon. The focus of this poster presentation will be on the fluorotelomer-based products of the PFAS group with

**WE075**

LIFE project PHOENIX: a new project for the management of water pollution from short chain perfluoralkyl acids in Veneto region (Italy)

F. Russo, M. Vazzoler, V. Groppi, Region Veneto, Direzione Prevenzione, sicurezza alimentare, veterinaria; F. Zanon, F. Da Prà, R. Lava, M. Mazzola, G. Onofrio, L. Da Rugina, ARPA Veneto; M. Bonato, University of Padua, Department of Biology and Department of Economics; F. Santovito, L. Tallandini, University of Padua, Department of Biology; M. Carrer, L. Palmeri, University of Padua / Department of Industrial Engineering; N. Tormen, University of Padua, Department of Biology; S. Valsecchi, F. Cappelli, Water Research Institute - Italian National Research Council IRSA-CNR; S. Polisello, Water Research Institute- CNR / Water Research Institute

In 2013 a significant episodes of PFAS pollution of surface and ground drinking water has been discovered in a large area of the Veneto region, in Northern Italy. The most important source of pollution was identified in a fluorochemical plant, sited in an area of groundwater recharge. The Veneto Region immediately put in place mitigation actions that were more effective for long chain PFAS than for short chain ones. For that reason, within the framework of the Community Life Program, the LIFE project PHOENIX: a new project for the management of water pollution from short chain perfluoralkyl acids in Veneto region (Italy), coordinated by the Department of Health Protection, Food and Veterinary Safety of the Veneto Region, in association with CNR IRSA, ARPAV and University of Padua, has been proposed and then funded. The activities of the LIFE-Phoenix project, acronym for"Perfluorinated compounds Holistic Environmental Institutional eXperience" started on 2017 and will end in 2020. LIFE PHOENIX project aims to show a new working method by including an Environmental Management System, supported through innovative forecasting tools based on ongoing monitoring, can manage risks related to the diffusion of persistent mobile organic contaminants (PMOC) such as short chain PFAS. This project will develop a set of institutional procedures and tools to assess and possibly prevent as well as respond to risks for environment and human health with the contribution of multidisciplinary specialists who will develop tools, protocols, guidelines and indications to assist authorities managing risks and emergency. The project will validate and compare some innovative technological tools for the mitigation of PFAS concentration in the water through a pilot plant adopting different techniques for the purification of irrigation water and drinking water, using full-scale plant (wetland system) and physico-chemical plans breakdown system (filters). The technologies applied to these experimental sites will be incorporated into an integrated management system that will serve as a model for managing analogous chemical pollution events from persistent and soluble polar substances.
The Groundwater of Hydrogeological Sub-Region II at Buenos Aires province, is well characterized from a physico-chemical perspective. Until today is well established that fluoride and arsenic are the main concerns related with the potential use as source of drinking water. However, an ecosystems perspective that focus on their invertebrate communities and the ecotoxicity potential is missing at least in this subregion. In this work we showed the preliminary results obtained after the sampling and analysis of ten on 20 total wells projected to be evaluated. This area is known as Junín Formation, which consists of sandy and silty sediments of silty loam to reddish brown to light brown color, very friable, and with scarce calcareous bodies of pedogenic origin. The Junín Formation of wind morphology constitutes an alternation of low elevations and depressions. Aeolian sediments, which belong to the Junin Formation (Aeolian Platense), normally do not exceed 5 m in thickness and rarely have calcareous (coarse) levels. The alluvial and colluvial deposits (sandy silts, sands, gravels and blocks) have a reduced vertical and areal expression. The samples were characterized according their main anionic and cationic constituents, presence of phyllosphate and chlorpyrifos, TOC, arsenic and fluoride. Also, cytogeneticity of concentrated waters were studied by comet assay using coelomocytes of Esenicia forida. Water quality was analyzed in combination with the dominant taxon of invertebrates founded. They were mainly epigean Copepods, Acari, Collembola, Insecta, Oligochaeta, Nematomorpha. A preliminary biotic and ecotoxic index were created to characterize each sampling well.

WE077 Chemical analysis, monitoring and toxicological evaluation of very polar compounds in drinking water and drinking water sources

R. Sjerp, KWR Watercycle Research Institute / Chemical Water Quality and Health; P. Kooij, KWR Watercycle Research Institute / K. Baken, KWR Watercycle Research Institute / CWG; A. Kolkman, KWR Watercycle Research Institute; P. de Voogt, University of Amsterdam / IBED

Very polar organic compounds are of special interest for drinking water utilities, since these substances end up in drinking water. The distribution of persistent and mobile compounds in the urban water cycle is widespread since they can leach to groundwater and/or pass wastewater and drinking water treatment. Currently there is an analytical gap, a monitoring gap and a lack of toxicity data for persistent and mobile organic compounds (PMOC). We aimed to close these gaps by the implementation of a target HILIC-MS screening method for very polar compounds and quaternary ammonium compounds and a nontarget HILIC screening. With these methods 45 samples from surface water, river bank filtrate, groundwater and drinking water in The Netherlands and Flanders have been analysed. Detected compounds include known contaminants melamine, uretroprin, metformin and guanylurea and newly detected compounds cyanuric acid and metformin. Despite the high removal rates during drinking water treatment (>70%), these compounds were found in drinking water. One compound is introduced during treatment: dichloroacetic acid. Most compounds and highest concentrations were detected in surface water and drinking water produced from surface water. The monitoring data of the very polar compounds gives insight into the seasonal variation of surface water quality. For the detected polar compounds human toxicological risk assessment is performed and results will be presented.

WE078 Beyond DEHP: High-molecular-weight phthalates and non-phthalate plasticizers in German Rivers

R. Nagotka, Federal Environment Agency (UBA) / Water and Soil; J. Koschorreck, Umweltbundesamt

The phthalate plasticizer Di(2-ethylhexyl) phthalate (DEHP) is a well-documented ubiquitous contaminant and frequently regarded as the predominant plasticizer in the environment. The use of DEHP and other phthalates was banned in the EU because of their endocrine-disrupting activity. As a result, the production of these phthalates decreased significantly. In contrast, the global plastics demand is continuously growing. Market data show that high-molecular-weight phthalates are now extensively substituting DEHP together with various types of non-phthalate plasticizers. Previous investigations on plasticizers in the aquatic environment have largely focused on DEHP and selected low-molecular-weight phthalates. However, little is known about the presence of other phthalates and non-phthalate plasticizers. In contrast, the objective of this study was to obtain spatio-temporal trends for DEHP and its substitutes in freshwater systems. We analyzed suspended particulate matter samples (SPM) for the presence of 23 plasticizers, i.e. 17 phthalates and 6 non-phthalates. Samples from recent years were obtained from the German Environmental Specimen Bank (ESB), which covered 16 sampling sites from major rivers including Rhine, Elbe and Danube. Retrospective trend monitoring with archived ESB samples that were collected over the last decade enabled trend analysis for DEHP and its non-regulated substitutes. Today, the high-molecular-weight plasticizer Disononyl phthalate (DINP) is the most abundant plasticizer detected in the SPM-samples. Our results indicate a fast appearance of new plasticizers like Dicapropylmononyl cyclohexane-1,2-dicarboxylate (DINCH) in freshwater environments and we identified several compounds, for instance Di(2-propylheptyl) phthalate (DPHP), as potential chemicals of emerging concern with increasing levels.

Wastewater effluents: How research can improve risk assessment and regulation (P)

WE079 Acute and chronic toxicity of Direct Blue 15 on microalgae and cladocerans: a comparative study

M. Hernández Zamora, Escuela Nacional de Ciencias Biologicas-I. P. N. / Laboratory of Experimental Hydrobiology; F. Martínez-Jerónimo, Escuela Nacional de Ciencias Biológicas-I. P. N. / Laboratory of Experimental Hydrobiology

Aquatonic pollution resulting from industrial activities, especially textile, leather, food and agrochemicals, is a major concern. Dyeing process of fabrics produces approximately 90% of the total textile wastewaters, containing significant concentrations of residual dyes. Colored wastewaters reduce light penetration in the water column, and affect photosynthesis of phytoplanktons. In addition, azo dyes are synthesized from carcinogenic compounds, such as benzidine; this can threaten the aquatic biota. The environmental impact caused by the discharge of textile dyes effluents has been scarcely studied; therefore, our study was aimed at evaluating the toxic effect of the azo dye Direct Blue 15 (DB15) on a primary producer (Pseudokirchneriella subcapitata) and on a primary consumer (Ceriodaphnia dubia). The microalgae was exposed to 4, 8, 16, 32 and 64 mg L\(^{-1}\) DB15 (96 h, 25°C, and continuous illumination of 120 \(\mu\)mol m\(^{-2}\) s\(^{-1}\)); the effects of DB15 on photosynthetic pigment and macromolecules content (proteins, carbohydrates and lipids) were assessed. The acute toxic effects of DB15 dye in cladocerans were determined at 48 h; tested concentrations were 100, 200, 400 and 500 mg L\(^{-1}\) at 25°C, 16.8 h photoperiod, with no food supply during the assays. In chronic toxicity tests C. dubia individuals were exposed to 5, 10, 15, 20 and 25 mg L\(^{-1}\) DB15 (7 days at 25°C, 16.8 h photoperiod, 1\(\times\)10\(^5\) cell mL\(^{-1}\) of P. subcapitata as food). P. subcapitata was more sensitive to DB15 (LC\(_{50}\) 13.0 mg L\(^{-1}\)) than C. dubia (LC\(_{50}\) 450 mg L\(^{-1}\)). Chlorophyll-a and -b were significantly increased in the algae exposed to all the dye concentrations, comparing with the control, but carotenoids were significantly reduced in all the DB15 concentrations. Concentration of proteins, carbohydrates and lipids per cell in P. subcapitata exposed to all DB15 concentrations were significantly higher than that measured in the control. In the highest 10 concentrations, total progeny, number of released clutches, and reproduction were significantly decreased in C. reducta; but age at first reproduction was significantly increased at 20 and 25 mg L\(^{-1}\) DB15. Results demonstrated that DB15 dye caused toxic effects of different magnitudes on aquatic biota (primary producer and primary consumer), for this reason, the azo dyes must be regulated to prevent environmental impacts caused by the discharge of textile dyes into waterbodies.

WE080 Integrated biomarker response calculation as a useful tool to assess the impact of effluents on the health status of fish

S. Wilhelm, University of Tuebingen / Animal Physiological Ecology; S. Jacob, Universität Tübingen / Animal Physiological Ecology; M. Ziegler, R. Triebekorn, University of Tuebingen / Animal Physiological Ecology

Wastewater treatment plants (WWTPs) are considered as one of the major sources of micropollutants in the aquatic environment. Many compounds have been linked to toxic and endocrine effects in aquatic organisms. The present study examines the impact of three WWTPs situated on different streams in Southern Germany on the health status of fish. Two of the investigated facilities are conventional treatment plants, combining mechanical, biological and chemical treatment. The third one was equipped with an additional powdered activated carbon filter unit, which has been in operation since October 2013. In order to examine the effects of the different effluents on fish, one-year-old rainbow trout (Oncorhynchus mykiss) were exposed in cages upstream and downstream of each WWTP effluent. Furthermore, the impact of the WWTP upgrade with activated carbon was investigated by comparing results of caging exposures conducted prior and subsequent to the upgrade. Several biomarkers, including histopathological alterations, the formation of micronuclei and binuclei, changes in vitellogenin levels, induction of hepatic ER\(_{\text{D}}\) activity, and changes in stress protein levels were examined, and the integrated biological responses (IBR) were calculated for the downstream exposure sites according to Sanchez et al. (2015), using the respective upstream site as a reference. IBR values for the conventional treatment plants (WWTP 1 and 2) differed slightly from each other, with WWTP 2 showing three to five times higher indices than WWTP 1. However, the highest IBR values were detected for male fish exposed downstream of the third WWTP prior to the upgrade with an activated carbon filter unit. After the installation of the additional treatment technology, a pronounced reduction of IBR values was observed. The IBR biomarkers proved to be a suitable approach to assess the impact of WWTP effluents on the health status of fish. Furthermore, it was a helpful tool to reveal the advantages of WWTP upgrading with powdered activated carbon.

WE081 Application of eco-genotoxicochemical and microbiological parameters for the assessment of the quality of wastewater industrial reuse

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Pharmaceutical residues in sewage effluents pollute the aquatic environment and may adversely affect fish populations. Conventional sewage treatment plants (STP) are not equipped to efficiently remove pharmaceuticals. Ozonation is emerging as a method to improve sewage treatment. Ozonation can however potentially create toxic by-products (OBP) that may have deleterious effects on fish. In this study we screened the concentrations of 103 pharmaceuticals and biological effects at a Swedish STP with a full scale parallel ozonation line. We investigated endocrine, reproductive and behavioral effects in fish and its progeny exposed to the conventional STP effluent. Furthermore, we sought to elucidate if any such effects were either abated or amplified by effluent ozonation (7 mg O₃/L). We exposed zebrafish to dechlorinated tap water (n=3), STP effluent (n=3) and ozonated STP effluent (n=5). The fish were exposed during 21 days (12:12 day/night cycle) in continuously replenished tanks (0.1 L/min, 25°C). During the last seven days, the fecundity (number of eggs produced per female) was measured and fertilized eggs were gathered from each replicate. The eggs were kept in our laboratory facilities and checked for mortality, malformations and locomotor activity at 6 days post fertilization. At the termination of the experiment, we recorded adult fish swimming activity and liver tissues was sampled for subsequent morphological and expression analysis. Results from the chemical screening showed that on average 77% of the screened pharmaceuticals were removed by ozone treatment. However, on the contrary to our assumptions, the biological effect screening revealed male larvae VTG-2 gene expression, a marker of estrogenic endocrine disruption, was induced by the ozonated effluent. This indicates that ozonation possibly created estrogenic OBPs. Furthermore, the ozonated effluent contaminated with organic and a tegens were more effective as an estrogenic inducer than the control. The possible genotoxic effect of wastewater using the Micronucleus test (MN-test). The study results showed a significant decrease in treated water samples of all microbiological parameters and the absence of E. coli. The ecotoxicological assays highlight a significant toxicity of the wastewater before the treatment while an evident decrease has been recorded after it. Sublethal effects for Danio rerio embryos and genotoxic effects for Vicia faba's micronuclei frequencies have been reported. Even though not yet conclusive, this approach can be considered a useful and promising tool in the reuse management of industrial wastewater and an initial support to the policy in this field.

**WE082 Comparative effects of the azo dye Congo Red on the green microalgae Ankistrodesmus falcatus and Scenedesmus incrassatus**

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**WE083**

**Exposure ozonation treatment: effects on adult zebrafish fecundity, behavior and vitellogenesis in a 21 day exposure study**

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weighted, homogenized and centrifuged for the determination of hydroperoxides, lipoperoxides, carbonated protein content, and the antioxidant activity of superoxide dismutase and catalase, results shown statistically significant increments regarding control group in all the biomarkers evaluated, thus indicates that the hospital effluent tested in this work can generate oxidative stress on Xenopus laevis larvae, based on the results obtained, hospital effluents can generate oxidative stress in other species and due the lack of appropriate WWTP hospital effluents can represent a risk for aquatic organisms.

WE086
An assessment of (anti)-androgenic activity in sludge from a rain spillway basin of the WWTP Aachen Soers as well as in sediments from the catchment area of the recipient water, the river Wurm
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Hormonally active micropollutants (MPs) are a well-known problem in aquatic environments. They can severely alter entire ecosystems by disrupting the endocrine system of its organisms. Adverse effects can extend to invertebrates and vertebrates including humans. As many MPs cannot be completely eliminated during conventional treatment in municipal wastewater treatment plants (WWTP), they enter the recipient waters. So far, mostly the effects of estrogenic active compounds have been investigated. However, a crucial part of the occurring hormone--equivalents is formed by androgen receptor inhibiting or activating compounds. Thus, the scientific attention has increased to account for their importance. Due to their strong lipophilicity, the main part of these compounds accumulates in sediments in sewage sludge. To measure endocrine-disrupting MPs is the implementation of an additional treatment step like ozonation. Within the DemOAC Project, the WWTP in the catchment area of the river Wurm in Aachen Germany will be equipped with a large-scale ozonation. It is unknown how the concentrations of (anti)-androgens in sediments will be altered by this new treatment step. Up to now, controversial results were published regarding the elimination of (anti)-androgens by ozonation in effluents. Furthermore, sediments were rarely studied. To fill this scientific gap, this study will evaluate the status quo of (anti)-androgenic potentials in sediments and samples from the WWTP before and after the implementation of the ozonation at the WWTP in 2018. Investigation of samples from a rain spillway basin was conducted to measure the potential endocrine impact in rural areas in heavy rain event. Additionally, a sewage sludge sample was tested, to gain more information. Assessment of (anti)-androgenic activity was performed by testing sample extracts using the (anti)-ARCALUX® assay. These studies will be conducted associated to the DemOAC Project as part of an exploratory study. First results revealed an (anti)-androgenic potential as well as cytotoxicity in the highest concentrations of the samples. The full depiction of the (anti)-androgenic activity in the catchment area of the Wurm will be available at the time of the conference.

WE087
Processes underlying the environmental fate of pharmaceuticals in the Nairobi River Basin “impact zone”: implication for environmental risk assessment
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Recent investigations have highlighted the widely-spread occurrence of active pharmaceutical ingredients (APIs) in African water bodies. Wastewater (treated or otherwise) is the main source of APIs to the environment and identified heavily contaminated areas have been ascribed to the poor African wastewater treatment facilities (WWT). The formation of so called “impact zones” is severe in urban areas characterized by informal settlements with little in the way of wastewater treatment. This study highlights how this is evidenced in Kenya and it is an example of such contaminated areas. The wastewater generated from the city’s informal settlements and the insufficient WWT is directly discharged in the Nairobi River basin leading to a large-scale “impact zone” characterized by occurrence of high levels of ammonia, biochemical oxygen demand and low dissolved oxygen and commensurately elevated levels of APIs. Also, the presence in this area of industrial wastewater discharges from pharmaceutical factories also contributes to the occurrence of APIs. Hitherto, the published investigations provided evidence of APIs occurrence at concerning concentrations, but little is known about the processes underlying the E-fate of such contaminants. Processes such as distribution to suspended solids, colloids and dissolved organic matter, the quality of these sorbents, and the ability of the biodegrading bacteriological community to adapt to and degrade a contaminant are key in determining its E-fate. The impact zone generated by sewage and industrial wastewater together with the local natural sorbents abundance influences the APIs distribution and E-fate. Data will be presented on sorbent concentration and quality related to distribution processes of APIs in the impact zone and the individualization of eventual bacteriological community shifts as an effect of the direct discharge of untreated wastewater loaded with APIs. In addition, since preliminary studies on the biodegradation of the antiretroviral nevirapine, commonly used in Africa, has showed persistency (similarly to studies on the antiepileptic carbamazepine in Western countries) the occurrence of nevirapine will be studied to test its suitability as indicator of sewage pollution in African river bodies. The implementation of this study contributes to the development of a methodology for the determination of the impact zone spatial boundaries and a dedicated environmental risk assessment approach for APIs in the impact zone.

WE088
Occurrence of pharmaceuticals, metabolites and transformation products from combined sewer overflows in London measured by high resolution targeted, suspect screening and untargeted chemical analysis
L. Barron, Kings College London / Analytical and Environmental Science; K. Munro, Kings College London; T.H. Miller, Kings College London / Analytical and Environmental Sciences; D.A. Cowan, Kings College London / Drug Control Centre; C. Martins, Therma Fisher Scientific; I. Perez, University of Aachen / Department of Biology Centre for Environmental and Marine Studies CESAM Combined sewer overflows (CSOs) are controlled releases of raw, untreated wastewater to a river during times of heavy rainfall to avoid back-flushing of buildings and streets. The impact of CSOs on a river catchment with respect to pharmaceutical residues is not well understood. In London, CSO occur ~12 times per week as its Victorian sewage network struggles to cope. Herein a temporal study of the River Thames is presented to identify CSO-related occurrence of pharmaceuticals, including metabolites and transformation products. Daily samples of river water, influent and effluent wastewater were analysed using a validated method involving solid phase extraction (SPE) and liquid chromatography and high resolution accurate mass spectrometry (LC-HRMS). The work was divided into four parts: (a) the identification of CSO markers based on their occurrence; (b) determination of 30 pharmaceutical/metabolite occurrences in influent and effluent wastewater; (c) screening for CSO markers receiving river water over a six-week period; (d) screening to identify metabolites/transformation products; and (d) classification of samples using untargeted data analysis. By differentiating influent and effluent, 10 CSO markers were identified including caffeine, bezafibrate, benzoylegone and furosemide which were present only at relatively high/consistent concentrations. Following this, targeted analysis of the River Thames samples revealed that CSO marker concentration increased mainly during wet periods where the tide was low. A further 14 compounds were also determined to observe any ‘dilution effects’ related to CSO release. In contrast to CECs, CSO concentrations are mainly influenced by population and civic activity. The potential occurrence of known metabolites/transformation products is also presented for a selection of compounds via machine learning prediction of LC retention times and mining of HRMS data [1]. Finally, untargeted analysis revealed that river samples could be differentiated based on climate and/or tide height using principal component analysis and volcano plots. The use of several different modes of data analysis provides a deeper understanding of complex occurrence data potentially influenced by timed CSO events. [1] K Munro, TH Miller, CPB Martins, AM Edge, DA Cowan, LP Barron, J. Chromatogr. A, 1396 (2015) 34-44

WE089
Occurrence, fate and bioactivity of pesticides in wastewater
V.V. Yargeau, McGill University / Chemical Engineering; P. Westlund, D. Nashogulu, S. Isazadeh, McGill University
Unlike for contaminants of emerging concern (CECs), data available for the occurrence, fate and bioactivity of pesticides (herbicides, fungicides, and insecticides) in wastewater treatment plants (WWTPs) is limited. Our research showed that of the 18 compounds investigated only imidacloprid, was not detected at the three WWTPs included in the study, confirming that municipal wastewater discharges contribute to the presence of pesticides in the aquatic environment. Using a suite of bioassays (high-throughput bioluminescence assay using the target species Vibrio fischeri, yeast estrogenic screen (YES) and yeast androgenic screen (YAS) using Candida albicans in pseudoinfection assay) as wastewater effluents treated using secondary treatment or ozonation was investigated. It was found that of the 12 pesticides showed either antiestrogenic or antiandrogenic activity and 7 compounds showed pleiotropic effects. This study was first to confirm endocrine activities based on yeast-based assays of recent neonicotinoids. The use of extended time points for the Vibrio fischeri, beyond the traditional 30 minutes, highlighted the potential longevity for some compounds was underestimated using only the acute test. Using a structure-activity relationship approach similar to the one used in hazard assessments, the relationship between toxicity and key physiochemical properties of the pesticides was investigated and trends were identified. This work also provided new knowledge on the removal of some fungicides (climbazole, myclobutanil and tebuconazole) by ozonation and demonstrated the recalcitrant nature of pesticides during ozonation. This study is one of the first to investigate androgenic activity during the ozonation of a mixture of pesticides and an increase was reported. These findings further demonstrate the importance of combining bioanalytical tools to analytical chemistry in the evaluation of wastewater quality.
WE090 Fate of perfluoroalkyl substances within a small stream food web affected by sewage effluent
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Within our experiment, the fate of fourteen PFASs was studied in an ecosystem of a small stream affected by SEPA’s effluent. The unique field experiment design was carried out to allow long-term study focused on bioaccumulation of PFASs in indicator organisms and their ability of adaptation to the polluted environment. Two hundred brown trouts (Salmo trutta) originating from clean site within the same stream were tagged and stocked downstream the source of pollution. Those fish were recaptured after one, three, and six months they spent in the environment affected by the effluent of that local STP. Besides the fish stocked into the polluted locality from the clean site, also fish originally inhabiting the downstream locality and macroinvertebrates from both sites were sampled and analysed. Passive sampling approach using polar organic chemical integrative samplers (POCIS) was applied to determine occurrence of PFASs in water soluble fraction over the course of the experiment instead of conventional grab water samples. Twelve of the fourteen target PFASs were found in concentration above the LOQ in at least one of the two sites, i.e., site from downstream locality while only three were present in samples from clean site. The compound pattern varied significantly between fish species of macroinvertebrates, and POCIS indicating several exposure pathways leading to bioaccumulation of PFASs in fish body. Concerning the accumulation of PFASs in fish, the highest concentrations were found in the liver of individuals sampled after three months of exposure. These concentrations rapidly decreased after six months although there was no significant change in occurrence of PFASs in water during the experiment. Such finding can be linked to both increase of water temperature leading to higher enzymatic activity and adaptation of studied fish to the polluted environment. Based on our results we also suggest that the process of adaptation might be related to the gender of fish as we found significantly higher accumulation of pollutants in males (first month of exposure). Acknowledgements: The study was financially supported by the Ministry of Education, Youth and Sports of the Czech Republic - projects “CENAKVA” (No. CZ.1.05/2.1.00/01.0024) and “CENAKVA II” (No. LO1205 under the NPU I program) and by the Ministry of Agriculture of the Czech Republic (NAZV “KUS” No. Q15350120).

WE091 Patterns of natural and human-made interacting processes on source, transport and fate of trace metals in the Adriatic Sea basin
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The Adriatic Sea has been under intensive influences of human activities, which are exhausting the marine ecosystems as a whole. The morphology of this land-locked marginal sea and the intensity of human pressure on its coastline influence enhances trace metal concentrations on sediments giving rise to pollution effects especially in the northern Adriatic Sea. This work was developed under the PERSEUS EU Project (Policy-oriented marine Environmental research in the Southern European Seas), guided by the Marine Strategy Framework Directive (MSFD), which aims to achieve Good Environmental Status until 2020 in European water bodies. Spatial and historical trends of sedimentary trace metals (Zn, Pb, Cu, Cr and Ni) and their characteristics (e.g., grain size and organic carbon) were assessed in recent layers and dated cores from western Adriatic Sea to reconstruct their sources, transport and accumulation. Our findings suggest that the Po River prodelta acts both as a bypassing and accumulation zone and exports ~30% of trace metals associated with the river gravel and planted with phragmites sp., carbon felt was used for the construction of anode and cathode, both with a surface of 0.72 cm². The distance between both electrodes was 6 cm without proton exchange membrane, both electrodes were connected by a resistance of 1000 Ohms. The feeding was performed by gravity applying four pulses of 1.5 L/h, using synthetic water whose composition was similar to date reported by Yadav et al. (2012). The effect of two types of contaminants was evaluated azo dye (AD) and alkylphenols (AP), the voltage measurements were made daily after 20 days of operation, the time necessary for the development of the biofilm (from 9.00 h in intervals of 2h). The
COD measurement was performed weekly on the influent and effluent of both systems. After 40 days in operation, the systems showed voltage average values of 673 and 580 mV, maximum current densities of 20.8 and 37.5 mA/m² and COD removal of 38.5 and 36.71% for effluents AZ and AP, respectively. A significant increase in the current density was observed in the measurements taken after 13:00 h, which shows an effect of temperature on the generation of voltage and therefore current flow in the system. The results obtained represent a sustainable option for the utilization of energy from domestic wastewater for secondary effluents, which would not only improve the quality of the water before being discharged to the receiving bodies but also take advantage of the high concentrations of nutrients contained in these wastes.

WE094 Adsorption of Crystal Violet from Quaternary Basic Dye Mixture onto A Sawdust-Based Adsorbent

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Wastewaters from textile and other dye-using industries usually contain more than one dye. In such an aqueous matrix therefore, the adsorptive characteristics of a dye species are likely to be affected by the presence of the other dye species in the mixture. The removal of crystal violet from aqueous solution by sulphuric acid treated sawdust (TAD) was investigated. The influence of malachite green, methylene blue and rhodamine B on its adsorption from binary, ternary, and quaternary dye systems were studied. The combined effect of mixture components and process parameters on the adsorption was studied and optimized using response surface methodology. The adsorbent was characterized and the experimental data obtained were fitted to different kinetics and isotherm models. The experimental results were further analyzed using Analysis of Variance (ANOVA) statistical test concept. The optimum contact time, pH, adsorbent dose and temperature were found to be 275.10 min, 9.94, 0.99 g and 60 °C respectively for the maximum decolourisation of 68.39 mg/L CV (97.2%). A linear model was obtained for the decolourization process through this design. The experimental values obtained were in good agreement with predicted values, and the model developed was highly significant, with correlation coefficient of 0.985. The adsorption in all the dye systems investigated followed Freundlich isotherm, and the maximum monolayer adsorption capacity was between 18.87 - 24.39 mg/g, depending on the composition of the adsorbate matrix. The adsorption kinetics was well described by the pseudo-second order model ($R^2 > 0.95$). All the eight adsorbate systems investigated were endothermic (ΔH positive; 35.30 to 43.60 kJ/mol), thermodynamically feasible ($ΔG^\circ$ 2.30 to -6.13 kJ/mol) and had increased entropy.

WE095 Diurnal patterns and removal of selected elements in two Norwegian wastewater treatment plants with primary treatment

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Discharges from wastewater treatment plants (WWTPs) are a source for pollutants to the environment. Here we studied influent patterns and removal of selected elements in two representative Norwegian WWTPs (HØRA and LARA) in Trondheim, Norway. Both WWTPs have significant industrial loading contributions, and removal of Al, P, S, Cr, Fe, Ni, Cu, Zn, As, Cd and Pb. To study release patterns and gain information about potential sources, diurnal variations of elemental concentrations were determined and modelled in 8-h composite samples of raw influent wastewater from morning, evening and night discharges. Element concentrations were highest in influent S (100% for Pb), Cd and Pb and lowest for Cd(< As>Cd and Pb). Concentrations of Al, P, Cr, Cu and Cd were higher in HØRA than LARA, with Fe loadings being approximately double. Removal efficiencies varied between the analysed elements, and were highest for Al (86%), Pb (74%) and Cu (57%) in LARA, which utilises both inorganic and organic flocculants. In contrast, removal rates were below < 50% for P, Cu and S in HØRA. However, in LARA, concentrations of Fe, Ni and S were significantly higher in the treated effluent compared to the raw influent, deriving from the use of inorganic flocculant. This was also reflected in Fe and S concentrations in treated sludge. Elemental concentrations in 8-h composite samples mostly followed general diurnal discharge patterns, with higher concentrations in mornings and evenings and lower concentrations at night. In HØRA, concentrations of most elements further correlates well with total suspended solid concentrations (TSS), with the strongest correlations observed for P, S and Cu ($R^2 > 0.9$). Correlations with TSS were less pronounced in LARA, and were weakest for Pb, Fe and Cu ($R^2 < 0.6$), which can be potentially attributed to the higher industrial loading contributions in LARA.

Enrichment factors were high for $\text{Pb, Cu, Zn, Cd, As}$, and were still above 10 for Cr and Ni in biosolids, indicating anthropogenic sources for these elements. Several elements also occurred as nano- and micron-sized particles.

WE096 Rapid detection of E. coli in wastewater effluent and impact of effluent discharge on riparian invertebrate diversity

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Effluent samples from wastewater treatment plants are expected, due to the treatment processes, to have lower concentrations of toxicants and pathogens than the influent. As such, they are expected to have a lesser impact on the environment than the influent. The aim of this study was to seasonally assess the efficiency of Phalaborwa’s wastewater treatment plant in removing pathogens ($E.\, coli$) from its effluent and to determine the impacts of that effluent on invertebrate diversity along the riparian zone of the receiving watershed. Loop mediated isothermal amplification technique was used to screen for the presence of $E.\, coli$ in effluent samples. There was negative identification of $E.\, coli$ in the samples collected. Impacts of effluent on invertebrates will be determined by analysing the changes in population dynamics of the invertebrates found in the riparian zone, above and below discharge point of the Wastewater Treatment Plant. At least, 13 morph species of eight insect families have been identified. Taxon diversity will be calculated using the Simpson-Yule Index, from where significant difference analysis would be calculated for environmental variable, species abundances and diversity data. More screening for the presence of $E.\, coli$ will determine the efficacy of Phalaborwa’s wastewater treatment plant in removing pathogens from its effluent. Further identification and presence or absence of invertebrates in the sampled riparian zone will determine if effluent has any impacts on invertebrate diversity.

WE097 The Demo3AC-project: Chemical and eco-toxicological investigations of the wastewater treatment plant Aachen

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Microplutonts (MPs) in municipal wastewater, like pharmaceuticals, are not sufficiently removed by conventional cleaning methods in wastewater treatment plants (WWTPs). As a consequence, complex mixtures of MPs discharged into the receiving streams and may cause various effects on the aquatic biota. To decrease the impact of MPs into the environment, a full-scale ozonation is implemented into the WWTP Aachen Soers, Germany within the Demo3AC-project. A crucial part of this project is the evaluation of the ecological and chemical state of the receiving stream, the River Wurm. This study focuses on the status quo of this river. To assess the impact of MPs, water samples were taken at 3 treatment steps within the WWTP and at 4 sampling sites (upstream and downstream the WWTP) along the river Wurm. The samples were subjected to high-performance liquid chromatography (HPLC) analysis for the detection of both native samples and extracts. To include various adverse effects a broad test battery was conducted (acute, chronic, mechanism-specific, in vivo and in situ) along with chemical analysis. The analysis of 60 MPs showed mainly the presence of pharmaceuticals and plasticisers/phthalates. In total 52 substances could be detected. A general elimination rate of about 55 % was determined. The toxic potential was high in the WWTP inlet for $D.\, magna$, $D.\, subspicatus$ and $D.\, rerio$. A. fisheri showed no effect. However, the toxicity was already markedly reduced at the outlet of secondary clarifiers within the WWTP. No acute and chronic toxicity was detected in any of the Wurm samples. Similar results were obtained in in situ feeding experiments with G. pulex. No significant differences in feeding rate between the sampling sites were recorded. On the other hand, significant mutagenic and endocrine effects were observed at the inlet and outlet of the WWTP as well as at all sampling sites within the Wurm. The $P.\, antipodarum$ reproduction assay showed also a significant increase in embryo production downstream of the WWTP. In parallel, experiments with river sediments and samples of an additional WWTP (upstream of the WWTP Soers) will be conducted to clarify to which amount the mutagenic and endocrine effects originates by diagenetic processes and as the WWTP Soers. The second part of this project will contain comparative studies investigating the situation after the implementation of the full-scale ozonation. This project is funded by the Ministry for Climate Protection, Environment, Agriculture, Conservation and Consumer Protection of North-Rhine-Westphalia.

WE098 To use or not to use: sewage overflow dredging

M.H. Wagemans, Bioclear earth

In a densely populated country like the Netherlands, with a dense sewage system,
many sewage overflows into surface water are present. Sediment located 250 m before and after the overflow needs to be discarded and burnt after dredging while 'normal' sediment can be reused as soil. Discord and burning is expensive for water boards, besides the fact that it costs a lot of energy and causes air pollution due to transportation, which is not sustainable. Also it causes a loss of raw materials. The water board Noorderzijlvest has started a pilot for reuse sewage overflow dredgings as soil for construction purposes for agriculture. For this purpose two depots have been set up: one with reference sediment and one with overflow sediment. During one year the concentrations of pharmaceuticals and pathogens will be monitored as well as antibiotic resistance and endocrine disruption. The basis of the project is the hypothesis that pathogens will be killed during the process of drying and pharmaceuticals will be (biologically) degraded which makes it possible to work in the depot. In June 2017 the sediments were sampled and analysed. In October dredging was performed and the pilots have been set up. The first monitoring round was performed one week after set up. In each monitoring round chemical analyses are performed on pharmaceuticals. Pathogens are identified by both culturing methods and Next Generation Sequencing (NGS), combined with viable PCR analyses to quantify specific pathogens that have been identified by NGS. Antibiotic resistance will be monitored by means of ESBI (extended Spectrum Beta-Lactamase) measurements. Endocrine disruption will be monitored by means of ER-Calux tests. In the presentation the background of the project will be sketched, then the results of the monitoring up till May 2018 will be presented. Also preliminary conclusions will be drawn and an outlook for reuse possibilities will be given as well as the meaning of the project for other water boards.

WE099 Assessing wastewater processes at oil refinery industry in Kazakhstan

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This paper presents the first attempt to assess the wastewater treatment processes at the oil refinery sector in Kazakhstan and evaluate to what extent these processes follow international and national regulations regarding industrial waste water treatment. The assessment was performed considering wastewater discharge from refinery processes at three factories in the country. While Kazakhstan's environmental regulation promote the polluter pay principle and follow WHO guidelines, oil refinery factories in Kazakhstan still exceed the permissible concentration of pollutants in discharged wastewater. The national regulation allows discharge of wastewater to natural or artificial ponds by not exceeding the concentration of pollutants in discharged wastewater. TPH concentration in wastewater is often exceeded at each sampling point due to the large size of the factory and the time gap between the start of pollution discharge by the factories and start of the environmental regulations. This leads to no incentive to treat wastewater in an efficient way. Additionally, the national law lacks regulations regarding detailed methodology to assess the pollutant substances in the discharged wastewater. Thus, the assessment by environmental authorities for each oil refinery is negotiated separately between the factory and the governmental body, giving the factory a strong position to define the parameters assessing the wastewater. As such, none of the factories provides analyses of, e.g., heavy metal contamination in discharged wastewater. TPH concentration in wastewater is often exceeded at each factory and there is no analysis done for different hydrocarbon fraction. Consequently, it is strongly recommended to provide a unified and transparent methodology for the country's oil refinery industry to assess all important pollutants in discharged wastewater and to include all types of hydrocarbon fractions.

Antibiotics and Antibiotic Resistance in the Environment: Fate and Ecological Effects, Resistance Development and Implications for Human Health (P)

WE100 Accumulation of Enrofloxacin in the sea lettuce Ulva lactuca

J. Rosa, University of Coimbra / Department of Life Sciences; S. Leston, CFE-Center For Functional Ecology / Department of Life Sciences University of Coimbra; A. Freitas, J. Barbosa, INIAV - Instituto Nacional de Investigação Agrária e Veterinária; M.F. Lemos, Instituto Politécnico de Leiria / MARE IPElieiria; M. Partida, CEF- Instituto de Ciências da vida / Departamento de Ciências da Vida University of Coimbra; F. Ramos, Faculty of Pharmacy University of Coimbra The demand for food products is pushing aquaculture to increase its production throughout the world. The increase in production can lead to negative effects since much more fish are growing in much smaller places. Aquaculture is still highly associated with the frequent use of chemical compounds in water, either to treat or prevent disease outbreaks in culture ponds. Integrated multitrophic aquaculture systems (IMTAs) can be a suitable approach to fish production, since one can have several species with different trophic levels growing together, where each species has its own economical value. Macroalgae can be used in such systems, usually at the exit point of the ponds, acting as biofilters and reducing the nutrient loading released to the environment. Contaminants such as antibiotics used in aquaculture can bioaccumulate in these organisms, passing to higher trophic levels. More recently, with the inclusion of macroalgae in human diets not only in Asia but also in other regions, we can ultimately ingest high levels of contaminants, which are not negotiated the same way for other fish products. Exposure tests were performed with the macroalgae Ulva lactuca in order to evaluate the effects of Enrofloxacin in growth. Antibiotic concentrations were measured in seawater and macroalgae discs at several sampling points, after immersion in an Enrofloxacin bath at two different concentrations. These results can help comprehend how IMTA systems can act in order to prevent contamination with antibiotics. As biofilters, these organisms are located at the exit point of fishponds or near cages, potentially accumulating pharmaceuticals.

WE101 Antibiotic resistance genes in manure, stored manure and soil after manure application

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Production animal farms are proposed to act as reservoirs where genetic material from microorganisms may be retained. After fertilization, the manure-associated ARGs and MGEs are present in soil, hence a consequence of fertilization. However, the abundance and number of these ARGs and MGEs clearly decreased from fertilized soil to unfertilized soil samples. Manure had the highest relative abundances of ARGs, and these manure-associated ARGs were not detected in unfertilized soil or ditch water sampled before fertilization. Likewise, ARGs abundant in unfertilized soil or in ditch water were not abundant or even detected in manure. After fertilization the manure-associated ARGs and MGEs were present in soil, consequently allowing discharge of high concentration of pollutants (total petroleum hydrocarbons (TPH) exceeds concentration by 30-80 times, ammonia (NH₃) by 25 times, total dissolved solids (TDS) by 6 times, biochemical oxygen demand (BOD) by 6 times and surfactants by 5 times) to pond. The reason for the initial high pond concentration is a result of a time gap between the start of pollution discharge by the factories and start of the environmental regulations. This leads to no incentive to treat wastewater in an efficient way. Additionally, the national law lacks regulations regarding detailed methodology to assess the pollutant substances in the discharged wastewater. Thus, the assessment by environmental authorities for each oil refinery is negotiated separately between the factory and the governmental body, giving the factory a strong position to define the parameters assessing the wastewater. As such, none of the factories provides analyses of, e.g., heavy metal contamination in discharged wastewater. TPH concentration in wastewater is often exceeded at each factory and there is no analysis done for different hydrocarbon fraction. Consequently, it is strongly recommended to provide a unified and transparent methodology for the country’s oil refinery industry to assess all important pollutants in discharged wastewater and to include all types of hydrocarbon fractions.

Antibiotics and Antibiotic Resistance in the Environment: Fate and Ecological Effects, Resistance Development and Implications for Human Health (P)

WE102 Bioaccumulation, biochemical responses and gene expression in the marine clam Scrobicularia plana exposed to a pharmaceutical mixture at sub-lethal concentrations

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Pharmaceuticals are pollutants of potential concern in the aquatic environment where they are commonly introduced as complex mixtures as a result of incomplete waste water treatment plant removal processes and improper disposal. Despite of pharmaceuticals occurring in the aquatic environments at trace levels (μg L⁻¹ to low μg L⁻¹), they have been specifically designed to be biologically active at low concentrations in human cells and therefore it is reasonable to assume that aquatic wildlife may also be susceptible to their effects particularly under condition of combined and chronic exposure. Hence the need to characterize biological effects in non-target organisms exposed to sub-lethal concentrations of pharmaceutical mixtures. Buprofen (IBU) is one of the most used non-steroidal anti-inflammatory drugs; its ability to induce toxic effects (i.e. oxidative stress, neurotoxicity, endocrine disruption, immunological alterations) in aquatic organisms at environmentally relevant concentrations has been widely proven. Ciprofloxacin (CIP) and fluoxetine (FL) are broad-spectrum antibiotics of the fluoroquinolones class. Fluoroquinolones toxicity was observed in rodents
producing among others oxidative stress, cyto, neuro and hepatotoxicity were induced after treatment with CIP in mice and rats. However, ecotoxicological effects on aquatic organisms of CIP and FL are practically unknown. In our study specimens of the clam *Scrobicularia plana* were exposed to control water and a mixture of IBU, CIP and FL at close environmentally relevant concentrations (10 y 100 μg/L each) during 21 days with the aim of studying toxicological responses allowing the assessment of the organism's life span or post-exposure depuration. Bioaccumulation of pharmaceuticals in clams was examined and changes in a suite of molecular biomarkers was used to evaluate the biochemical status of clams during both exposure and depuration: biochemical responses related to oxidative stress (CAT, SOD, GR, T-GPx activities and LPO levels), detoxification (GST activity) and neurotoxicity (AChE activity), and mRNA expression of genes associated with regulatory system of xenobiotic exposure. Results obtained indicate a general activation of oxidative stress and neurotoxicity related features in enzymatic responses as well as changes in genetic profiles suggesting that selected pharmaceutical mixture in aquatic environment represent a risk for the clam *S. plana*.

**WE103**

Changes in the environmental risk of veterinary antibiotics after the introduction of antibiotics-reducing policies

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In recent decades, pharmaceuticals in the environment have been concerns for environmental protection. Especially in the livestock sector, the residual antibiotics in the environment could lead to adverse effects on non-target organisms, contamination of food and drinking water supplies, and increased antimicrobial resistance (AMR). Since 2000s, in Korea, the policies for reducing antibiotics usage, such as Separation of Dispensing and Prescribing of Drug, or Restriction of Adding Antibiotics in Animal Feed, have been enforced in view of public health management. Those policies have reduced the antibiotics usage since the enforcement of these policies, the assessment for these policies in view of environmental risk has not been performed yet. In our previous work (1), an emission prediction model for calculating the predicted environmental concentrations (PECs) of the active pharmaceutical ingredients (APIs) used not only for human but for veterinary purposes was presented. For veterinary usage, the model covers pharmaceutical life cycle posterior to consumption, including direct discharge, manure composting, and land application over the agricultural soil. The emission model was combined with SimpleBox and SimpleTreat into a single spreadsheet-type model for calculating the PECs. In the present study, antibiotics which are of top 26s in veterinary consumption in each year from 2001 to 2016 were selected as target substances. In this period, the environmental risk posed by the regulated antibiotics (Tetracycline, Norfloxacin, etc) has decreased, however, the use of non-regulated antibiotics (Florfenicol, Tilmicosin, etc) have steeply increased (255 times for Florfenicol, 13 times for Tilmicosin), implying emerging risk. The method used in the present work may serve as a quantitative tool to efficiently assess the policy about pharmaceuticals concerning their environmental risk. Reference (1) Eun Jeong Han; Dong Soo Lee, Application of emission prediction model for calculating the predicted environmental concentrations for antibiotics in Korea, Annual Meeting of Society of Environmental Toxicology and Chemistry, Brussels, Belgium, 2017

**WE104**

Development of microplate based assay and its application to establish differences in cyanobacteria sensitivity to antibiotics

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Antibiotics are continuously entering the environment and pose a risk to environmental health through their direct effects upon microorganism communities and to human health through antimicrobial resistance. They are designed to target bacteria but in current environmental risk assessment (ERA) only one species of cyanobacteria and the activated sludge respiration inhibition test (ASRIT) are proven not to be sensitive for antibiotics) are used to represent all bacterial diversity. There is therefore concern that the potential impacts of antibiotics on environmental health are not fully considered in ERA. We have developed a microtitre assay that broadly follows and meets the validity criteria of the OECD 201 test guideline as a cost effective way to determine the effect of antibiotics on cyanobacteria growth. We applied this assay to determine growth-rate effects on 8 species of cyanobacteria and total antibiotics to establish differences in species sensitivity for the improvement of the ERA of antibiotics. Our key findings are: 1) the performance of the microtitre assay is suitable for accurate and reliable assessment of effects on growth inhibition in a wide range of bacterial species; 2) differences in cyanobacteria sensitivity to antibiotics can span several orders of magnitude; and 3) the current framework for ERA of antibiotics inadequately addresses the risk to bacterial populations and testing several diverse cyanobacteria species will increase confidence in the protection goals established.

**WE105**

Direct and indirect effects of antibiotics in the leaf-shredding macroinvertebrate Gammarus fossarum

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Recent studies indicated that both leaf-decomposing microorganisms (i.e., bacteria and fungi) and macroinvertebrate detritivores (i.e., shredders) can be affected by antibiotics via direct and indirect pathways (i.e., via altered microorganism-mediated food quality). However, relatively little is known about these effects on shredders. Therefore, we performed a comprehensive study involving three experiments, which aimed at unravelling the importance of waterborne and diet-related effects of the antibiotic ciprofloxacin (CIP) on the model shredder *Gammarus fossarum*. During a 7-day feeding activity assay, we assessed the effects of waterborne CIP exposure on gammarids’ survival and feeding activity, while alterations in leaf palatability for *G. fossarum* due to microbial colonization of the leaves in the presence of the antibiotic were investigated using food choice assays (i.e., diet-related pathway). Furthermore, during a long term assay of 24 days, sublethal effects (the shredders’ energy processing and physiological fitness) were assessed when either subjected to a waterborne or diet-related exposure to CIP. Our assays showed that increase in aminoglycoside and sulfonamide resistances, while an increase in aminoglycoside and sulfonamide resistances occurred in a few aminoglycoside and sulfonamide resistances occurred in a few isolates. The temporal evolution of the resistances shows that there is a marked reduction (> 97%) in the genes coding for tetracycline, b-lactam, and aminoglycoside resistance, while an increase in aminoglycoside and sulfonamide resistance genes were observed. These genes usually form part of integrins, which have more persistence into the environment. Besides, we have found positive correlations among almost all ribosomal protection genes and with the deactivation genes, whereas off pump genes were positively correlated among them, suggesting that the persistence of antimicrobial resistance genes could be related to their mechanisms of action. In conclusion, although the composting process does not end up eliminating the AR genes, it can be considered a alternative to the environmental management of the avian manure. RTA2014-00012- C03-02 and S2013/ABI-2747.

**WE107**

Environmental Assessment Of Multi-Class Pharmaceutical Residues In the Tejo Estuary

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Evaluating the use of veterinary antibiotics in dairy environments

RTA2014

Evaluating the use of veterinary antibiotics in dairy environments indicated to minimize the risk of these drugs. This work is funded by De la Torre et al., 2012. The environmental relevance of these results is discussed included: the avian density and the capacity of the soil to accumulate this antibiotic organisms, specifically in plants (RQ> 1). No risk is identified for CIPR. Finally, an addition method (MultiCriteria Decission) and two risk factors were implemented for each matrix are being applied for the detection and quantification. The knowledge gathered will then be applied to antibiotic resistance studies using the pharmaceuticals detected at the highest concentrations and in mixtures. The knowledge gathered will then be applied to antibiotic resistance studies using the pharmaceuticals detected at the highest concentrations.

WE108

Environmental risk of enrofloxacin used in aviculture

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The presence of veterinary antibacterials residues in animal excreta supposes a health and environmental hazard associated with its agricultural reuse. Many of them have toxic potential for terrestrial and/or aquatic organisms. The environment can act as a reservoir not only for residues, but also for antimicrobial resistance genes, and may spread them into the food chain. This is a particularly serious in the case of antibiotics that can accumulate in soil, such as fluoroquinolones, which have a high adsorption capacity forces, high bioavailability, and high persistence. This adds of this work focuses on the environmental risk assessment of enrofloxacin (ENR) and its main metabolite (ciprofloxacin, CIPR), associated with its use in poultry farming in Spain according to the technical prescriptions of the authorized products. The environmental risk ratios (RQ) have been calculated following the European Guidelines on Environmental Risk Assessment of Veterinary Drugs (EMEA/CVMP/ERA/418282/2005). In case of the CIPR, information has been used in the metabolism and excretion of the ENR in chickens, to estimate the levels of CIPR in soil and later, to assess their environmental risk. The results indicates that the estimated PECsoil for ENR (443 μg/kg), implies risk for terrestrial organisms, specifically in plants (RQ= 1). No risk is identified for CIPR. Finally, an ENR environmental risk map has been generated in Spain. Allowing us to identify the "hot spots" where the greatest environmental management and surveillance efforts should be applied. This spatial analysis (ArcGIS 10.2) was carried out using a simple addition method (MultiCriteria Decision) and two risk factors were included: the avian density and the capacity of the soil to accumulate this antibiotic (De la Torre et al., 2012). The environmental relevance of these results is discussed and the effectiveness of methods and strategies in preventing ciprofloxacin in battery cages is indicated to minimize the risk of these drugs. This work is funded by RTA2014-00012- C03-02 and S2013/ABI-2747.

WE109

Evaluating the use of veterinary antibiotics in dairy environments to inform on environmental risk assessment approaches

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The University of Nottingham owns a high throughput dairy farm with around 2000 milking cows, from which the processed liquid waste ends up in a 3000 m³ slurry tank. Contained within the slurry we can find antimicrobial foot washings, waste milk and bedding particulates (among other components). Large volumes of liquid slurry are produced in farms and this waste material is commonly applied to fields as a fertilizer in the UK. The health of a dairy herd is supported by the administration of antibiotics so the dairy farm setting works as an antimicrobial resistance (AMR) reservoir and as a route of entry into the environment. Antibiotics’ persistence in dairy settings occurs through mediums such as slurry, milk and subsequently, in soil via spreading. These routes can act as a channel for the transference of compounds and the insenmation of AMR within the food chain through intake by plants and migration to other sources via water run-off, possibly affecting human and animal health against human and animal health. Therefore, the increasing soil retention of polar substances after soil amendment and the high persistence found for some antibiotics in batch experiments (e.g. sulfonamides), make further research on exposure assessment necessary along with the analysis of veterinary antibiotics in dairy environments in order to assist in shedding light on the long established concern of the environmental fate and behaviour of veterinary antibiotics in farming and to propose better handling practices as a basis for future regulations. The main objective of this study is to measure the distribution of select antibiotics currently in use within the farm environment, relating their presence to the length of use and the last date of prescription. For this purpose, specific optimized analytical methods including 19 antibiotics belonging to 10 different families were employed for clean-up and pre-concentration of different matrices (slurry,soil,milk) by means of liquid-liquid extraction, followed by solid-phase extraction (SPE) and identification and quantification by liquid chromatography-mass spectrometry (LCMS). For instance, several veterinary antibiotics under study were detected in wastewater slurry with concentrations up to 18.6 μg L⁻¹ for oxytetracycline suggesting a high number of treatments. Summarizing, this is the first step towards an evaluation of the impact of antibiotics presence and fate in the AMR development.

WE110

How do marine and freshwater cyanobacteria react to long term exposure of antibiotics? Is there a potential for increasing antibiotic resistance in the environment?

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An increasing amount of pharmaceuticals are detected in waterbodies all over the world. Antibiotics are of special concern for the environment: Due to the high amount of medication, its specific activity against prokaryotes and a possible antibiotic resistance formation in the environment. Cyanobacteria have prokaryotic target structures of antibiotics and are of high importance for the primary production and nitrogen cycle in marine waters. To investigate the long term effects of antibiotic exposure on limnic and marine cyanobacteria, the limnic cyanobacterial test according to OECD 201 and the marine cyanobacteria test described by Heseding and Floeter in 2016 were performed. Exposed cultures were recultured at the end of the test and then reexposed to the same antibiotic active substance as part of a repeated test. As test organisms Synechococcus leopoliensis (limnic cyanobacteria) and Synechycystis spec. (marine cyanobacteria) were selected. The tests were carried out on a 24-well microwell plate. In the test, the percentage inhibition of the growth rate (cell number) is determined in comparison to the negative control over a period of 72 hours. The derived EC50-values after repeated exposure were compared for different antibiotics of environmental concern. The investigations are carried out as part of the PharmCycle project.

WE111

Impact of antibiotics on the feeding rate of the freshwater shrimp Gammarus pulex

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Antibiotics are one of the main categories of pharmaceuticals and their release into the freshwater environment can impact the natural bacterial and fungal communities, which can threaten the survival of organisms that rely on them as a food source. One such organism is the freshwater detritivore Gammarus pulex that commonly feeds on detritus such as, naturally conditioned Alnus glutinosa leaves. The study aim was to establish if the feeding rate of Gammarus pulex was altered when their food source (Alnus glutinosa) was exposed to environmentally realistic concentrations of antibiotics during the natural leaf conditioning process. An investigation included three antibiotic scenarios (1) exposure to the bacteriostatic agent Tetracycline, (2) exposure to a mixture of Sulfamethoxazole and Trimethoprim bacteriostatic agents that are commonly prescribed together and (3) exposure to the bactericidal agent Broad-spectrum antibiotic Ciprofloxacin. 24 h leaf discs were performed using Alnus glutinosa leaf discs of 1.3 cm Ø and standardised Gammarus pulex specimens (n=60). The organisms were kept at 15°C under a 12:12 h light:dark cycle. 15 replicates were undertaken with three environmental realistic concentrations and a charcoal filtered tap water control (200 mg/L, 20 mg/L and 2 mg/L). The leaf discs were photographed at the start and finish of the investigation and these images underwent analysis with Image J software in order to calculate the area consumed. After 24 h, the Gammarus pulex were sacrificed by exposure to −20°C temperature before being dried at 60°C for 24 h and weighed. This protocol was performed with antibiotic scenario 1, 2 and 3. The results showed that Tetracycline (Z=0.198, p=0.897) and Ciprofloxacin (Z=1.568, p=0.120) showed no significant effect on the feeding rate of Gammarus pulex. 24 h exposure to Substantially lower than expected amounts of Sulfamethoxazole (Z=-2.189, p=0.029) were observed on the feeding rate of Gammarus pulex, indicating an inhibition of the feeding rate. The results indicate that the presence of environmental antibiotics can result in a reduction of the feeding rate of Gammarus pulex.
p=0.667) were not a concern in relation to feeding at environmentally realistic concentrations (scenario 1 and 3), (p<0.05). When exposed to a mixture of Sulfamethoxazole and Trimethoprim (scenario 2) there was an impact on the Gammarus pulex feeding rate (Z=13.239, p<0.004). However, further investigation would be required to investigate these drugs individually to identify if the obtained results were driven by one or the combination, and also to establish if there is a genuine environmental concern associated to this mixture or if the data is blurred in some way.

WE112 Persistence of the sulfamethoxazole antibiotic in a digestate-amended agricultural soil


Thousands of tons of antibiotics are annually used in human and veterinary medicine worldwide. They are excreted, from the treated organism, either unaltered or as metabolites, reaching soil and water ecosystems. In particular, the use of wastewater, sewage sludge, livestock manure and digestate as agricultural amendments and fertilizers, introduce residual concentrations of antibiotics to soils. Livestock raising practice involves the use of antibiotics in feed; consequently digestate obtained by anaerobic digestion of manure may be an additional source of antibiotics and resistance genes in soil. Sulfamethoxazole (SMX) is one of the most commonly prescribed and consumed sulfonamide antibiotics, due to its ability to inhibit Gram-positive and Gram-negative bacteria it is used in veterinary practices, agriculture and livestock breeding both for treating diseases and promoting growth. However, current knowledge about its persistence and possible environmental effects is poorly understood. In the present study, we investigated the persistence and the possible effects on the soil natural microbial community of SMX in an agricultural soil amended with solid anaerobic digestate from bovine manure anaerobic fermentation. Microcosms, containing soil and digestate treated with 20 mg/kg of SMX, were set up in the presence/absence (sterilized soil) of the natural microbial community. Moreover, non-antibiotic-treated microcosms were used as microbiological controls. At fixed times (0d, 7d, 13d, 20d, 61d), SMX residual concentrations (ASE extraction and HPLC-UV detection) and microbiological parameters (cell viability, abundance and activity) were analysed. Finally, a molecular marker for antibiotic resistance (intI1 gene (class 1 integron) was investigated at the start and the end of the experiment using qPCR. Results showed that although an acute negative effect (0d) was observed on the microbial abundance and viability, the antibiotic was degraded in just a few days. Interestingly, the intI1 gene was found in the soil where the digestate was added, showing its introduction through this agricultural practice.

WE113 Pollution in the Mooi River: Fluconazole and fluconazole resistant pathogenic yeasts species

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The presence of yeast species in water sources that are associated with faecal pollution have been demonstrated. Some of the isolated species are potentially pathogenic and could cause superficial and life-threatening infections. Fluconazole on the other hand is the most used antifungal agent in the treatment of yeast infections as well as prophylactic agent to prevent Candida and Cryptococcus infection in HIV patients. The aim of the study was to determine yeast pollution, susceptibility of these to, and the levels of, fluconazole in the Mooi River, North West (South Africa). Yeast isolates were enumerated using membrane filtration, selective media and incubation at 37°C and identified using biochemical methods (staining and 26S rRNA gene sequencing). Resistance to fluconazole was determined by disc diffusion. Environmental DNA was isolated directly from water using membrane filtration and a commercial DNA isolation kit. Yeast levels as indicated by qPCR were determined. Yeast levels were determined for fluorescence analysis, water samples were extracted using solid phase extraction. The extracts were analysed with liquid chromatography coupled to a quadrupole time-of-flight mass spectrometer. The purified isolates identified included Candida albicans, C. krusei, C. tropicalis and Saccharomyces cerevisiae. The yeasts identified have been associated with polluted waters. Some isolates in the present study are pathogenic and have direct contact with polluted water could cause infections to immune compromised people and were all resistant to fluconazole. Quantitative PCR of the 26S rRNA gene indicated that a high number of gene copies were present at all sites. Fluconazole levels ranged from

WE114 Reactivity, mobility and degradation of the antibiotic Sulfamethoxazole and its impact on the microbial communities of an agricultural soil amended with organic waste products

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The fate of Sulfamethoxazole (SMX), a sulfonamide antibiotic widely spread in natural soils and waters, was evaluated in batches and columns of a silty-loam soil under culture from Feucheralsen (INRA Versailles, France). SMX revealed to be a weak sorbent since in batches only 10% of SMX sorbed at a solid/solution ratio of 0.1. It’s sorption increased strongly with soil organic matter content (addition of manure), indicating that the essential feature of OM addition is an increase in sorption sites density at almost constant sorption strength, and confirms previous results about the strong influence of sorbed complexants such as Cu(II). The mobility of SMX evaluated in water-saturated columns showed higher mobility of SMX than expected from the sorption study. Unexpectedly, this increased mobility was increased by the soil, suggesting mobile SMX contributes to antibiotics transport in soils. Batch degradation experiments revealed that SMX removal is quite fast with half-life values ranging between 18 and 350 days in non-sterile and sterile soils. This degradation process was shown to occur principally in both the coarsest and finest soil-size fractions, while almost no biodegradation was observed in the mass-dominant silty fraction of the soil in agreement with its low microbial biomass content. The impact of SMX on the soil bacterial community, evaluated through total biomass (16SDNA), qPCR (Antibiotic Resistance Genes, ARG, sul1 and sul2), DGGE fingerprinting and high throughput sequencing revealed important impacts of SMX on soil microbial biodiversity and species richness and the emergence of specific taxons, resistant to the antibiotic. These results permitted to characterize the global fate and impact of SMX in an agricultural soil. SMX appeared to be mobile in presence of added OM. SMX appeared also quite readily biodegradable, especially when in contact with coarse and fine soil size fractions, where it had the strongest impact on soil bacteria. Keywords: Antibiotics, SMX, organic matter, impact, DGGE, Miseq, ARB, biodegradation

WE115 Risk assessment of antibiotic resistance and related genes in human impacted environments

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The origins of antibiotic resistance in the environment is relevant to human health because of the increasing importance of zoonotic diseases as well as the need for predicting emerging resistant pathogens. Antibiotics are used in diverse settings for food production. Domestic animals are treated with antibiotics for both curing disease and promoting growth. Aquaculture is also used to manage infectious disease. Wastewater treatment plants receive sewage from various sources, including hospitals and households which are both important sources of antibiotics and their residues, and antibiotic resistant bacteria. Risk assessment of antibiotic resistance is complicated. It should include at least quantitative information of the gene, sequence of the gene, host cell of the gene and gene function. Moreover, due to the complexity of antibiotic resistance, non-targeting humans and animals. Antibiotics in particular have played a decisive role due to their ability to inhibit growth or eliminate microorganisms. Unfortunately, its misuse combined with bacterial capability to acquire antibiotic resistant genes, have significantly contributed to the escalation of life-threatening
infections leading to worldwide antimicrobial resistance (AMR). This issue is most evident in artificial high selective pressure settings (e.g. hospitals, animal farms) but the increasing occurrence of antibiotics and resistance genes in the environment is spawning serious concern. The measured concentrations of antibiotics are relatively low, most are readily biodegradable and there are considerable resistance-associated fitness costs. However, intricate bacterial compensatory mechanisms, population dynamics and long-term persistence can lead to resistance gene emergence and enrichment (e.g. via horizontal gene transfer). Therefore, there is a need for a better understanding of how concentrations of antibiotic relate to the abundance of resistance genes in different environmental compartments under different conditions. In this study, we compiled this sparse information by conducting an extensive literature meta-analysis to evaluate global trends. Our investigation showed that the environmental matrices under investigation differ in terms of microbial abundance (e.g. surface water). Interestingly, there are cases where gene variation is weakly correlated with antibiotic concentration (e.g. sediment) which challenges the common proportionality assumption between these two parameters. This indicates that AMR genes can be highly maintained throughout bacterial communities under certain environmental conditions. Whether detected gene levels are antibiotic-induced or the consequence of changes in microbial communities (e.g. via urban effluents) is still under debate. These results are expected to support the development of integrative models capable of providing meaningful risk assessment to support decision-making.

**WE117**

**Sulfamethoxazole degradation in river water microcosms and effect on the natural microbial community**


The widespread use of antibiotics causes concern on their occurrence and fate in different environmental matrices. Following administration, antibiotics are partially metabolized and a large amount is excreted unaltered or as active metabolites, reaching wastewater treatment plants (WWTW). Most WWTWs are not able to remove them efficiently and through their effluents, they reach surface water. The synthetic compound sulfamethoxazole (SMX) is one of the most prescribed and consumed sulphonamide antibiotics to treat urinary tract infections used both in human and veterinary practices. The widespread use of SMX implies its antibiotic-induction or the consequence of changes in the environmental matrix. Furthermore, SMX is not readily biodegradable and resistant to hydrolysis. The high frequency of detection and relative persistence of SMX in environment cause a potential risk of antibacterial resistance spread in ecosystems. Multiple mechanisms confer sulfonamide resistance in bacteria, although data on biodegradation and spread of antibiotic resistance genes (ARGs) in natural water ecosystem are quite scarce. The aim of the present work was to investigate the SMX degradation in natural river water in presence/absence of the microbial community and to identify the occurrence of sul genes associated to the antibiotic resistance. Microcosm experiments were set up using river water treated with 500 µg/L of SMX. At fixed times, water sample were collected for chemical (SMX residual concentrations) and microbial analysis. The disappearance time of 50% of the initial SMX concentration (DT50) and the effects of the microbial community on the environmental matrix were measured. For microbial communities, results were compared to large reference environments. The study demonstrates the potential for antibiotics to be transferred from inland areas into larger water bodies. Antibiotics entering streams can arise from various sources. In urban areas, antibiotics of human and veterinary origin can enter streams due to runoff or leaching from surrounding areas, but most notably from wastewater discharges that release effluent directly into streams. In rural areas, antibiotics can enter streams from application in the maintenance of livestock, which due to runoff and leaching, can contribute to veterinary antibiotics being present in rural streams and groundwater. Work from the present study found human and veterinary antibiotics in both urban and rural streams. Antibiotics detected include sulfamethoxazole, sulferamazene, trimethoprime, danofloxacin, sulfaquinoxaline, streptomycin, enrofloxacin and tylosin, with several of the detected antibiotics being present in both urban and rural streams. Results from this work demonstrate that streams are key sites regulating discharge of antibiotics to larger bodies of water and that surrounding land use and infrastructure influences the presence of antibiotics in streams in urban and rural areas.

**WE120**

**The Role of Water Quality Analysis: Understanding our process environment to inform on AMR**

T.P. Dodsworth. The University of Nottingham / Biosciences; R. Hellwell, The University of Nottingham / Social Sciences; E. King, The University of Nottingham / Biosciences; R.L. Gomes, The University of Nottingham / Faculty of Engineering Politicale delle Marche; M. Magro, E. Bonaiuto, F. Vianello, G. Radaelli, A.D. Gray, University of North Carolina at Greensboro / Biology; D. Todd, University of North Carolina at Greensboro / Chemistry; A.E. Hershley, University of North Carolina at Greensboro / Department of Biology.

Antibiotic pollution is of concern to environmental health sciences due to the implications associated with their presence in the environment. There is a great deal of literature drawing attention to antibiotics in the environment contributing to antibacterial resistance amongst bacteria. Antibiotic pollution in stream ecosystems in the United States remains a research area that has received little attention, while more research has focused on antibiotics in larger rivers, lakes and estuaries. Streams ecosystems play a crucial role in the transport of water from inland areas to coastal waters as well as to rivers and lakes, and have greatest potential to mitigate the occurrence of antibiotic pollution in larger systems. The presence of antibiotic microorganisms in streams demonstrates the potential for antibiotics to be transferred from inland areas into larger water bodies. Antibiotics entering streams can arise from various sources. In urban areas, antibiotics of human and veterinary origin can enter streams due to runoff or leaching from surrounding areas, but most notably from wastewater discharges that release effluent directly into streams. In rural areas, antibiotics can enter streams from application in the maintenance of livestock, which due to runoff and leaching, can contribute to veterinary antibiotics being present in rural streams and groundwater. Work from the present study found human and veterinary antibiotics in both urban and rural streams. Antibiotics detected include sulfamethoxazole, sulferamazene, trimethoprime, danofloxacin, sulfaquinoxaline, streptomycin, enrofloxacin and tylosin, with several of the detected antibiotics being present in both urban and rural streams. Results from this work demonstrate that streams are key sites regulating discharge of antibiotics to larger bodies of water and that surrounding land use and infrastructure influences the presence of antibiotics in streams in urban and rural areas.

**WE118**

**The effect of antibiotics on representatives of aquatic algal and plant species**

B. Cerrina, Politecnica delle Marche; M. Magro, E. Bonaiuto, F. Vianello, G. Radaelli.

Antibiotics contamination of the environment is a significant portion of the pharmaceuticals that are in use in the country. However, antibiotics as environmental contaminants have received little attention in Kazakhstan and the topic is a new field for research in the country. The aim of the present study was to the impact of priority antibiotics in use in Kazakhstan on representative aquatic species. *Lemna minor* and *Chlorella sp.* were selected for the ecotoxicological investigations. Five major use antibiotics in Kazakhstan (amoxicillin, chloramphenicol, azithromycin, sulfamethoxazole, oxytetracyclines) and their mixture were used in the experimental assessments. The compounds were selected based on a previous prioritization study based on the risks of active pharmaceutical chemicals (APIs) to aquatic environments in Kazakhstan.

The study on *Lemna minor* was conducted according to the OECD Guidelines for the testing of chemicals 221. *Lemna minor* species were cultured in Swedish Standard (SIS) growth medium and effects of the antibiotics on growth assessed over 7 days. The results of the study showed EC50 values of each test compounds ranged from 2.8 to 21.8 mg/L. *Lemna minor* was most sensitive to the sulfamethoxazole, with its EC50 being below 10 mg/L. The test on algae was conducted according to the OECD Guidelines for the testing of chemicals 201. *Chlorella* were cultured in Tamano medium and algae numbers were counted in Goryaev chamber under a microscope. The macrocide substances azithromycin and chloromycin were found to be the most toxic compounds to the algae with EC50 values being lower than 1 mg/L. In the future, it is recommended to perform assessments on the sensitivity of other less well studied aquatic species to priority AMRs in Kazakhstan as well as monitoring studies to establish levels of exposure in the country. This will then provide a basis for the risk of these substances to be established.

**WE119**

**The Presence of Human and Veterinary Antibiotics in Urban and Rural Streams of North Carolina**

A.D. Gray, University of North Carolina at Greensboro / Biology; D. Todd, University of North Carolina at Greensboro / Chemistry; A.E. Hershley, University of North Carolina at Greensboro / Department of Biology.

Antibiotic pollution is of concern to environmental health sciences due to the implications associated with their presence in the environment. There is a great deal of literature drawing attention to antibiotics in the environment contributing to antibacterial resistance amongst bacteria. Antibiotic pollution in stream ecosystems in the United States remains a research area that has received little attention, while more research has focused on antibiotics in larger rivers, lakes and estuaries. Streams ecosystems play a crucial role in the transport of water from inland areas to coastal waters as well as to rivers and lakes, and have greatest potential to mitigate the occurrence of antibiotic pollution in larger systems. The presence of antibiotic microorganisms in streams demonstrates the potential for antibiotics to be transferred from inland areas into larger water bodies. Antibiotics entering streams can arise from various sources. In urban areas, antibiotics of human and veterinary origin can enter streams due to runoff or leaching from surrounding areas, but most notably from wastewater discharges that release effluent directly into streams. In rural areas, antibiotics can enter streams from application in the maintenance of livestock, which due to runoff and leaching, can contribute to veterinary antibiotics being present in rural streams and groundwater. Work from the present study found human and veterinary antibiotics in both urban and rural streams. Antibiotics detected include sulfamethoxazole, sulferamazene, trimethoprime, danofloxacin, sulfaquinoxaline, streptomycin, enrofloxacin and tylosin, with several of the detected antibiotics being present in both urban and rural streams. Results from this work demonstrate that streams are key sites regulating discharge of antibiotics to larger bodies of water and that surrounding land use and infrastructure influences the presence of antibiotics in streams in urban and rural areas.

**WE121**

**Safety and efficiency assessment of antibiotic administration by magnetic nanoparticles in Zebrafish**


Antibiotic resistance is a significant problem in human medicine and can also impact the environment. Antibiotics can be released into the environment through various sources such as sewage treatment plants, runoff from agricultural fields or animal waste, and leaching from soil. This can lead to the development of antibiotic-resistant bacteria, which can persist in the environment for long periods of time. The presence of antibiotic-resistant bacteria can pose a threat to human and environmental health, as they can spread resistance genes to other bacteria that are not naturally resistant. Therefore, it is important to understand the sources and mechanisms of antibiotic resistance in the environment.

In this study, we investigated the fate and effects of antibiotics administered to zebrafish using magnetic nanoparticles. The aim was to assess the safety and efficiency of this method of antibiotic administration. The nanoparticles were designed to target specific organs or tissues, allowing for targeted delivery of antibiotics. The study was conducted using a well-characterized zebrafish model, which allows for detailed monitoring of antibiotic distribution and resistance development.

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The indiscriminate use of antibiotics in the aquaculture sector has raised public concern because of possible toxic effects, development of bacterial resistance, and accumulation of residues in individual tissues. Even if several countries have developed regulations about their use, it is clear that long-term growth of the aquaculture industry requires both ecologically sound practices and sustainable resource management. Alternative strategies for better management of antibiotic administration are of primary interest to improve antibiotic use rates and, as a consequence, to reduce their release into the aquatic environment. The present study investigates, for the first time to our knowledge, a new methodology for oxytetracycline (OTC) administration through the use of iron oxide nanoparticles (NPs) (made of maghemite γ-Fe₂O₃) in zebrasfish (Danio rerio). These were divided into 4 experimental groups: control; group A exposed to 4µg/L OTC (through weekly 10µg of OTC and 100µg of bupivacaine hydrochloride); group B exposed to 4µg/L OTC; and group C exposed to bare NPs. HPLC analysis, histological analysis and other methods were applied to perform different evaluations. No detoxification processes or anatomical alterations were observed in fish exposed to bare NPs. Exposure of fish to the SAMNs@OTC complex resulted in a 10 times higher OTC accumulation with respect to using water exposure. This new method for OTC administration seems more efficient with respect to the traditional way of exposure and shows the potentiality to reduce antibiotic utilization and possible environmental impacts.

Analysis and Fate of Emerging Contaminants in soils, water and plants under water scarcity (P)

WE122 Identifying and Controlling Sources of Ultra-Trace Metals in Control Blanks and Ensuring High-Quality Data for Sensitive Environmental Risk-Based Decisions

R. Vitale, Environmental Standards; L. Dupes, A. Reed, Environmental Standards Inc; M. Mc Anulty, BP Exploration

The control of blank contamination is a necessary requirement when evaluating the quantitative and qualitative accuracy of analytical results for decision-making purposes of site delineation, toxicological evaluations, and site remediation. This control is essential to differentiate actual site concentrations from potential sources of contamination. Contaminating elements can be inherited from background site metals conditions and cleanup criteria. Trace-level concentrations of total and dissolved metals were observed in several equipment rinse blank samples collected at multiple sites during the field sample collection season of 2014 through 2016. The detection of total and dissolved metals in these field quality control (QC) samples resulted in qualification of both total and dissolved sample results resulting in questions regarding the presence or absence of low-level site contamination. Although equipment rinse blank samples do not have specific contamination acceptance criteria due to the field collection process, the goal of equipment rinse blanks is to verify that contamination was not introduced during the sample collection process or by sampling equipment. The purpose of the investigation and identification was to determine potential sources of metals contamination in equipment rinse blanks that can be identified, reduced, or eliminated. Multiple avenues of potential contamination were investigated including a study of sample tubing, peristaltic pumps and in-line filters; field observation of equipment blank collection processes; initial and post water-quality monitoring; sample bottle cleanliness and storage of deionized water for use in collection of the equipment rinse blank. This presentation will provide details of the investigation process and resulting implementation of several important corrective actions.

WE123 Comprehensive Analysis of Elemental Contamination in Environmental Samples by Polarographic Single Pore License Plating ICP-MS (ICP-MS)


The analysis of elemental contamination is a significant task for laboratories working in environmental analysis. Besides direct regulation of contaminant levels, for example in waters, also a number of other sample types must be screened, such as sludges or sludges. Targeted elements comprise the "big four", arsenic, cadmium, mercury and lead, but also many other elements. Special challenges in this area include the need to often measure large numbers of samples and potential complications caused by a variety of spectral interferences. For example, if rare earth elements are present in a given sample, they can lead to severe interferences on arsenic and selenium. To avoid false positive results, triple quadrupole ICP-MS is ideally suited for effectively removing all potential interferences. However, the inherently higher complexity of a triple quadrupole based system in comparison to traditionally applied single quad systems is a barrier for most laboratories dealing with routine analysis. This presentation will highlight the use of ICP-MS, especially triple quadrupole ICP-MS, for the analysis of environmental samples. Dedicated software solutions, such as tools to simplify method development, increase productivity or tackle advanced applications, such as chromatographic sample pretreatment or sample introduction, will be presented to show the broadness of available applications using modern ICP-MS instrumentation.

WE125 ANALYTICAL METHOD FOR DETERMINATION OF FULLERENE (C60) NANOPARTICLES IN SEAWATER SAMPLES

L.R. Diniz, Universidade Estadual do Maranhão / Agroecology. L. Capellini, Universidade Federal de São Paulo UNIFESP / Departamento de Química

Fullerenes are allotrope forms of carbon produced in highly energetic processes of carbon origin or anthropogenic sources. In the last years, the increasing application of nanomaterials in several areas of human endeavor besides their physical and chemical properties, contribute for the growth of the global economy. However, the growing production and application of nanomaterials is also promoting discussions about the possible risks of these compounds to the environment and human health. Data have already been reported on the occurrence of fullerenes in different matrices, including the atmosphere, soils and sediments, and fresh water. Despite this, little information has been related to marine environments while coastal areas and estuaries are suspected to be one of their major sinks. The purpose of this study is developed and optimize an analytical method to evaluate the presence of nanomaterial fullerene (C60) in seawater samples. It will be tested two methods of extraction, that are the liquid-liquid micro extraction (DLLME), and (2) QuEChERS, after, all the samples will be analyzed by liquid chromatography tandem mass spectrometry (LC-MS/MS). Keywords: Marine pollution. Fullerenes. Nanomaterials.

WE126 Screening of per- and polyfluoroalkyl substances (PFASs) and total organic fluorine in wastewater effluent from Nordic countries

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The discharge of per- and polyfluoroalkyl substances (PFASs) into the environment via wastewater is a pressing public health issue. Apart from the frequently detected PFASs, such as PFOS and PFOA, more and more novel PFASs have been reported recently. We used the target screening method to identify novel and legacy PFASs in the Nordic environment. The aim of this project is to determine as many PFASs and total organic fluorine as possible in wastewater effluents from the Nordic countries and self-governing areas, including Finland, Sweden, Norway, Denmark, Faroe Islands, Iceland and Greenland. Field collected effluent (250 mL) were extracted by solid phase extraction (SPE). Identification and quantification of target compounds was performed by use of liquid chromatography tandem mass spectrometry (LC-ESI-MS/MS), ultra-performance convergence chromatography (UPLC®) and combustion ion chromatography (CIC). The significance of the occurrence, levels and patterns of various PFASs in Nordic wastewater effluents are discussed.

WE127 Quantitative evaluation of lag effect in polar organic chemical integrative sampler (POCIS) and modified POCIS with polytetrafluoroethylen (PTFE) membranes

Y. Jeong, H. Kwon, KIST Europe / Environmental Safety Group; H. Jeon, KIST Europe; A. Meyer, E. Furufrock, H. Beck, Saarland University, K. Smith, RWTH Aachen University / Institute for Environmental Research; A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics

Increasing occurrence of organic contaminants in the aquatic environment has heightened the need for reliable and efficient monitoring technique. Traditional grab sampling followed by laboratory extraction and instrumental analysis is well established method, but only provides a snapshot of the water quality status. Achieving representative sample with grab sampling takes considerable labour, time and cost. Here, time-integrative passive sampling technique is recognized as a promising monitoring tool. Passive sampling technique allows the simple sampler construction and application, provision of time weighted average concentration and in situ sampling. Various configuration of passive sampling devices are currently available, one of which is polar organic chemical integrative sampler (POCIS). POCIS consists of Oasis HLB® solid phase extraction (SPE) PVTFE) with polytetrafluoroethylene (PTFE) membranes and has been widely used for the detection of hydrophobic contaminants in the past decade. However, uncertainties in quantitation of POCIS measurements have been pointed out as a main limitation of POCIS. Compound specific sampling rate depends on sampler configuration and environmental parameters such as flow rate and temperature. Lag effect from membrane sorption within POCIS further complicates the transfer kinetics of analyte. In this study, modified POCIS (POCIS-PTFE) with polytetrafluoroethylene (PTFE) membrane instead of PES membrane was tested in an attempt to avoid or lower the PES membrane sorption. The primary aim of this study is to (1) introduce modified POCIS and (2) identify the membrane sorption within POCIS in order to better understand partitioning kinetics of POCIS. In the laboratory experiment, the analyte mass fraction in membrane relative to total POCIS (i.e., Oasis HLB plus membranes) and membrane-water partition coefficient were determined for a range of compounds (log KOC from -0.03 to 6.26). Less membrane sorption was found in
in water, pore water, and sediment samples from Baiyangdian Lake, China. A total of 31 water samples and 29 sediment samples were collected in March, 2017. Caffeine was detected with 100% frequency in surface water, pore water, and sediment samples. Carbamazepine was detected with 100% frequency in surface water and sediment samples. Five N-APs were prominent, with mean concentrations of 4.90–266.24 ng/l in surface water and 5.07–14.73 pg/g in sediment samples. Four M-APs were prominent, with mean concentrations of 0.97–29.92 ng/g in pore water samples. The total concentrations of the different classes of PPCPs followed the order: N-APs (53.26%) > M-APs (25.39%) > SAs (10.06%) > TCs (7.64%) > QNs (3.64%) in surface water; N-APs (42.70%) > MCs (25.43%) > TCs (14.69%) > SAs (13.90%) > QNs (3.24%) in sediment samples, and MCs (42.12%) > N-APs (34.80%) > SAs (11.71%) > TCs (7.48%) > QNs (3.86%) in pore water samples. The geographical differences of PPCP concentrations were largely due to anthropogenic activities. Sewage discharged from Baoding City and human activities around Baiyangdian Lake were the main sources of PPCPs in the lake. An environmental risk assessment for the upper quartile concentration was undertaken using calculated risk quotients, and indicated a low or medium high risk from 18 PPCPs in Baiyangdian Lake and its five upstream rivers.

**WE131** Occurrence of perfluorinated compounds in air, water, soil, sediment, and fishes from the Asan Lake region, South Korea

J. Lee, Y. Lee, J. Lee, Seoul National University; S. Kim, Eulji University; M. Kim, Seoul National University / Department of Health Science; Y. Kho, Eulji University; K. Zoh, Seoul National University / Department of Environmental Health

Perfluorinated compounds (PFCs) are known to be endocrine disrupting chemicals and can cause adverse effects on human health and environment. In July and October 2017, ambient air (n=2), fresh water (n=24), sediment (n=24), soil (n=24) and freshwater fish (n=27) samples were collected in Asan lake region, and the levels of six PFCs in samples were measured. The levels of TCS and PFOS were detected with freshwater between Pyeongtaek and Asan cities in Gyeonggi Province, and provides water for nearby industrial complex and agricultural areas. Two large streams join the lake and there are many industrial complexes near the streams. To analyze 16 PFCs, 2 samples were taken at each stream and 8 samples were taken at main lake. Analyses were carried out using LC/MS/MS after solid phase extraction. The results showed that concentrations of ∑PFCs were ND–19.6 pg/g, ND–447.9 pg/g, ND–9.7 mg/g dry weight (dw) in surface, pore and sediment waters, respectively. PFOS, PFHxS, PFDS and PFBA were detected in all water, sediment, and fish samples. In air, 85% of total PFCs mainly existed as the gas phase compared to particulate phase. In water and sediment, higher levels of some PFCs were observed at the confluence of two streams, implying that anthropogenic activities from industrial complex in the upstream are the main source of contamination. Our study first reported the data of 16 PFCs levels in multimedia environment including air, water, soil, sediment, and fishes in Korea.

**WE132** Seasonal changes in water and sediments’ microplastics in a Mexican estuary (Tecolutla).

L. Arques Hernández, P. Ramirez Romo, U.A.M. Iztapalapa / Hidrobiología Microplastics (MP) are persistent contaminants that measure less than 5 mm, they have additives that are vectors of other POPs and metals, which can cause deleterious effects to the organisms that ingest them. MP can increase the temperature and decrease the sediments permeability. On the other hand, plastic particles are efficiently transported through water. There are just a few MP studies in Mexican aquatic ecosystems, so the objective of the present work was to evaluate the seasonal changes in numbers, size, color and form of the MP present in water and sediment of Tecolutla’s estuary. Water and sediment samples were collected in five different sites in three different climate seasons (northern storms, dry and rainy). In the laboratory water volume was measured and filtered through a 0.45 μm filter, which was later dried at 50 °C for 24 h. Sediments were dried at 50 °C, a 40 g subsample was taken and hydrogen peroxide (30 %) was added to disintegrate all organic matter, followed by a zinc chloride solution (pH = 1.5) to float the MP particles. Later the solution was decanted through a cellulose filter and dried at 50 °C for 24 h. MP particles on the filters were observed through a microscope (Dissecting followed by digital Celestron) and photographs of the particles were obtained to have information on the polymer identity. The polymer identity was done through Scanning electron microscopy. MP size in water range from 10 to 1,730 μm, and their presence was higher in the northern storms season, followed by dry and rainy seasons with the highest numbers in a small tributary. Sediments had more MP and higher numbers in the dry season, with size ranging from 30 to 2,500 μm with highest numbers in the boat dock. Black was the most abundant color in both matrices followed by blue and red. Most MP were fibers. In conclusion MP were present in water and sediments year round, bigger particles were found in sediments and smaller in water. This is the first evaluation of MP in a Mexican estuary so continuing this type of research is important to

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**WE128** Occurrence and Ecological Risk Assessment of Several Endocrine Disrupting Chemicals in Urban River Water and Sediment of South China

C. Huang, Jinan University; L. Wu, Y. Guo, Jinan University / School of Environment

This study mainly focused on the occurrence, distribution, and ecological risk assessment of eight selected endocrine disrupting chemicals (bisphenol analogues, parabens, and triclosan) in urban river water and sediment of south China. The eight target chemicals were detected in both water and sediment samples with concentrations ranged from not detected to 66500 ng/L and from not detected to 492 mg/g dw, respectively. Among this eight chemicals, the top three major chemicals were bisphenol A (BPA) (account for 35%), methyl paraben (MeP) (23%), and triclosan (TCS) (14%) in water, while BPA (43%), TCS (37%), and MeP (14%) in sediment. Significant correlations were found between most of the selected EDCs, especially MeP and TCS both in water and sediment (p < 0.01), indicating that these chemicals were in common sources and widely usage. After calibration, our solid phase extraction (SPE) 13.1 μl injections of target substances were flowed into Liuxi river annually based on the 89 primary stream. The ecological risk assessment showed that TCS was the most dangerous compound to aquatic organisms with average HQ = 1.57 (up to 11.5) in river water and average HQ = 0.74 (up to 3.36) in sediment. And the possible joint toxic effect of selected chemicals showed that aquatic organisms were severely exposed to diverse EDCs.

This study suggested that compared to the main rivers, the endocrine disrupting chemicals in streams deserves more attention.

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**WE129** Occurrence, distribution and fate of pharmaceuticals as chemical markers of contamination from urban sources in the vulnerable area of the Ebro Delta (Spain)

M. Celis, M. Gros, Catalan Institute for Water Research ICRA; M. Farre, IDAEA CSIC Barcelona; D. Barceló, M. Petrovic, Catalan Institute for Water Research ICRA

The Ebro Delta and the upper part of the Ebro basin have widely been exposed to contamination from both agriculture and urban sources, being wastewater treatment plant effluent discharges the dominant contamination path. In order to determine the distribution and impact of contamination from urban sources in the vulnerable area of the Ebro Delta, water and sediment samples were collected at different sites, covering reaches of the Ebro river located upstream and downstream WWTs, lagoons, irrigation channels and coastal areas. Water samples were analyzed using ultra-high performance liquid chromatography coupled to tandem mass spectrometry, using a hybrid triple quadrupole–linear ion trap instrument (UPLC–QqLIT–MS/MS). In order to assess seasonal variations, distribution and fate of pharmaceuticals, 16 sampling campaigns in 2016 were performed, covering autumn, winter and spring-summer. Fifty seven and thirty one out of 81 pharmaceuticals were found in water and sediment samples, respectively. Analgesics/ant-inflammatory, lipid regulators, cholesterol lowering statin drugs and antibiotics were the most frequently detected pharmaceuticals, with the highest concentration found in river water, while the lowest concentration were found in sea water. The occurrences of pharmaceuticals detected in sediment samples showed lower frequency of detection than in water. Nevertheless, some compounds were only found in sediments, and not in water, such as the synthetic glucocorticoid (dexamethasone), the antiabetic (glibenclamide) and the diuretic (furosemide). Salicylic acid was the most ubiquitous quantified compound in sediment samples, with a maximum concentration of 18.2 ng g⁻¹ dw (furosemide). Salicylic acid was the most ubiquitous quantified compound in sediment samples, and widely usage. After calculation, our solid phase extraction (SPE) 13.1 μl injections of target substances were flowed into Liuxi river annually based on the 89 primary stream. The ecological risk assessment showed that TCS was the most dangerous compound to aquatic organisms with average HQ = 1.57 (up to 11.5) in river water and average HQ = 0.74 (up to 3.36) in sediment. And the possible joint toxic effect of selected chemicals showed that aquatic organisms were severely exposed to diverse EDCs. This study suggested that compared to the main rivers, the endocrine disrupting chemicals in streams deserves more attention.
understand the biological significance of their presence.

WE133
Simultaneous biodegradation of water treatment additives: Transformation byproduct formation, impact of biode, shock dosing and salinity
T. Wagner, University of Amsterdam / IBED; J. Parsons, University of Amsterdam / IBED-ELD; A. Langenhoff, H. Rijnsdarts, Wageningen University / Environmental Technology; P. de Voogt, University of Amsterdam / IBED
Securing the supply of fresh water to fulfill the demand of the rising world population is identified as one of the largest environmental challenges in the near future. The Water Nexus research program aims at developing integral solutions for problems with water scarcity in delta areas worldwide. A significant fraction of industrial fresh water is taken up in on-site cooling towers. Several treatment technologies such as reverse osmosis, electrodialysis and membrane distillation may facilitate the reuse of discharged brackish cooling tower water. However, cooling towers water contains different water treatment chemicals such as corrosion inhibitors, biocides and antiscalants that hamper the optimal functioning of the treatment technologies by, for instance, membrane fouling. An interesting water pre-treatment option is the use of CW systems such as constructed wetlands (CWs). Biodegradation is one of the main contaminant removal mechanisms in CWs. However, the biodegradation potential of CWs for many of the water treatment chemicals is not well understood. In this study, the simultaneous biodegradation of different representative water treatment chemicals by bacteria from CWs was explored. The representative water treatment chemicals comprised: 

- benzotriazole (corrosion inhibitor), DNPB (biocide),
- glutaraldehyde (biocide), PEG (surfactant) and HEDP (antiscalent). The following questions are addressed: Does shock dosing with biocides affect the CW biodegradation potential for the target chemicals? What is the influence of different salinities on the biodegradation of the target chemicals? Which signature microbial transformation products are being produced by single target chemicals that can be used to monitor the fate of the CW systems? How can we use the results of the salt tracer tests and provide information about hydrological loss and gain for the Schönbrunnen River. Dilution, mixing and dispersion processes can be identified with tracer tests and determine the residence time available for pollutant transformation. The quantification of the mass transport of pollutants in the river is possible by analysing conservative ions. This helps to derive and characterize chemical processes like photo-degradation, sorption to particles or biochemical processes in biofilms from target screening data, mainly on pesticides. With the collected information from different disciplines, we get a larger picture about the pollutant mass transport in the Schönbrunnen River and adjoining compartments.

WE134
Fate of organic micropollutants in a small river: hydrological and chemical processes
C. Glaser, Center for Applied Geosciences / Center of Applied Geoscience; M.E. Müller, Eberhard Karls Universität Tübingen; F. Faltermeier, Eberhard Karls Universität Tübingen / Center of Applied Geoscience; C. Zwiener, Environmental Analytical Chemistry, Center for Applied Geoscience, University of Tübingen / Geosciences; M. Schwientosk, Eberhard Karls Universität Tübingen; C. Zarifi, University of Tübingen / Center for Applied Geoscience
Knowledge on pollutant transformation from laboratory experiments often fails to describe observations in the field. Thus, the CRC-CAMPUS aims to describe the fate and metabolism of anthropogenic pollutants on the landscape scale in different compartments in the Ammer catchment. This study is part of the sub-project ‘Rivers’ and will identify and quantify the dominant processes from hydrology and chemistry which influence the fate of organic micropollutants in river systems. Field investigations take place in the Schönbrunnen River close to Tübingen (Germany) in the southwest of Germany, which is mainly influenced by agriculture. Salt tracer tests are combined with measurements of conservative ions and chemical tracers to determine the processes occurring in the Schönbrunnen River. The results are based on the results of the salt tracer tests and provide information about hydrological loss and gain for the Schönbrunnen River. Dilution, mixing and dispersion processes can be identified with tracer tests and determine the residence time available for pollutant transformation. The quantification of the mass transport of pollutants in the river is possible by analysing conservative ions. This helps to derive and characterize chemical processes like photodegradation, sorption to particles or biochemical processes in biofilms from target screening data, mainly on pesticides. With the collected information from different disciplines, we get a larger picture about the pollutant mass transport in the Schönbrunnen River and adjoining compartments.

WE135
Occurrence of pharmaceuticals at extremely high concentrations in surface waters in Nigeria
O.M. Ogunbawo, University of Leeds / Geography (Physical); P. Kay, University of Leeds / School of Geography; L. Brown, University of Leeds / School of Geography (Physical); J. Wilkinson, The University of York / Natural and Built Environments; A. Soares, Universidade Federal de Santa Catarina / Biochemistry Department; C. Sinclair, The Food and Environment Research Agency / Centre for Chemical Safety and Stewardship; R. Shabi, Lagos State Environmental Protection Agency
Pharmaceutical pollution of surface waters is increasingly recognized as a global problem, but to date, there have been no detailed studies from most African countries. In this study, the occurrence of 37 pharmaceuticals belonging to 19 therapeutic classes was studied in surface water and effluent in Lagos State, Southwest Nigeria. Samples were collected year-round from 22 surface water sites, and 27 compounds were detected at least once, many at extremely high concentrations. Maximum concentrations for a range of compounds, including trimethoprim, sulftamethoxazole, cimetidine, atenolol, and paracetamol were in the order of 150 microg L\(^{-1}\). The mean concentrations for sulftamethoxazole, trimethoprim, cimetidine, paracetamol, lidocaine, metformin, carbamazepine and atenolol were 55.90 microg L\(^{-1}\), 38.69 microg L\(^{-1}\), 31.62 microg L\(^{-1}\), 24.99 microg L\(^{-1}\), 22.55 microg L\(^{-1}\), 20.98 microg L\(^{-1}\), 15.35 microg L\(^{-1}\), and 15.10 microg L\(^{-1}\) respectively. Vanlaxafine has the lowest mean of 4.231 ng L\(^{-1}\) other than the 10 compounds listed. Those not detected. When compared with published data from around the world, these values are several orders of magnitude higher than most studies of pharmaceutical occurrence but similar to some other peak concentrations measured in developing countries such as China and India. Seasonal variations were observed for certain pharmaceuticals, i.e., antibiotics, paracetamol, tramadol, metformin, lidocaine, and carbamazepine which may be related to the endocannabinoid system.

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**WE136**
Assessment of emerging contaminants in the L’Albufera Natural Park (Valencia, Spain)
D. Sadutto, University of Valencia / Environmental and Food Research Group, CIDF (GV, UV, CSIC); M. Andrés Costa, Universitat de València / Environmental and Food Safety Research Group; R. Alvarez-Ruiz, University of Valencia; Y. Pico, University of Valencia / Medicine Preventive

Metals play a critical role in maintaining natural cycles and supporting a wide range of biodiversity. They regulate water quantity, groundwater recharge, and can contribute to regulating floods and the impacts of storms, and provide the fish and rice. The Albufera Natural Park is one of the most representative and valuable coastal wetlands in the Valencia Region and the Mediterranean basin. Covering an area of 125 hectares, which has increased the quality of life per capita due to the growing population (>1600000 inhabitants) has introduced a number of emerging contaminants that threaten this wetland. In this study, 42 drugs of abuse and 45 pharmaceuticals have been studied in influents and effluents of 10 Wastewater Treatment Plants (WWT), 7 irrigation channels and the Lake of L’Albufera de Valencia. Ten different conditions of the influents were analyzed, and sewage treatment plants were characterized. This study aims to evaluate the water quality of the Iacorbni river in estuarine region, in order to evaluate the antrhopic changes occurring in the surroundings. Three sites were chosen, in which sediments and water samples were collected. The sediments were analyzed for the presence of sterols and pharmaceuticals by GC / TOF-MS after extraction with methyl tert-butyl ether. The water samples were analyzed with respect to the parameters: 

- ammonia concentration, total phosphate, total phosphons, fecal coliforms and sulfide, according to the methodologies described in Standard Methods (APHA). TOF-MS chromatographic analyzes of sediments and water samples were also performed on extracts obtained using SPE (Strata-X/dichromethane). The results obtained showed high concentrations of ammonia, nitrates and total phosphate, besides high fecal coliforms. Between the analyzed sterols, cholesterol and derivatives such as coprostanol were identified at varying concentrations in the sediments of the several sites. Estradiol derivatives and drugs such as anxiolytics and remedies for sleep control were present in GC / TOF/MS chromatographic analyzes. The results confirmed the high contamination of the waters of the Iacorbni River by the discharge of domestic sewage. Ecotoxicological tests using fish are being conducted, including assay to assess genotoxicity.

### References

- Sadutto, D.; de Voogt, P.; Rijnaarts, H.; Langenhoff, A.; Alvarez-Ruiz, R.; Soares, M.; Sadutto, D.; de Voogt, P.; Rijnaarts, H.; Langenhoff, A.; Alvarez-Ruiz, R.; Soares, M. The metropolitan region of Florianopolis has undergone an intense urbanization process in recent years, which has modified the landscape and the quality of life in this region. The objective of the present study was to evaluate the water quality of the Iacorbni river in estuarine region, in order to evaluate the antrhopic changes occurring in the surroundings. Three sites were chosen, in which sediments and water samples were collected. The sediments were analyzed for the presence of sterols and pharmaceuticals by GC / TOF-MS after extraction with methyl tert-butyl ether. The water samples were analyzed with respect to the parameters: 

- ammonia concentration, total phosphate, total phosphons, fecal coliforms and sulfide, according to the methodologies described in Standard Methods (APHA). TOF-MS chromatographic analyzes of sediments and water samples were also performed on extracts obtained using SPE (Strata-X/dichromethane). The results obtained showed high concentrations of ammonia, nitrates and total phosphate, besides high fecal coliforms. Between the analyzed sterols, cholesterol and derivatives such as coprostanol were identified at varying concentrations in the sediments of the several sites. Estradiol derivatives and drugs such as anxiolytics and remedies for sleep control were present in GC / TOF/MS chromatographic analyzes. The results confirmed the high contamination of the waters of the Iacorbni River by the discharge of domestic sewage. Ecotoxicological tests using fish are being conducted, including assay to assess genotoxicity.

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The wastewater generated by the human activity contains several anthropogenic contaminants, including emerging pollutants. Sewage sludge is generated during the dephosphoration process in the wastewater treatment plants (WWTPs). The study of emerging pollutants present in this sludge are far limited comparing with the water, mainly because of the challenge that involve their high content in organic matter making difficult their handling, storage and analysis. In Spain, the 80% of this sludge comes from the agricultural sector as fertiliser, and the presence of pollutants could affect to the surrounding ecosystems. The sludge samples are from different treatment plants next to the Natural Park of the Albufera in Valencia, an area surrounded by 14000 hectares devoted to rice crops. Samples were extracted by liquid phase extraction (LPE) with mixture of EDTA-McIlvaine buffer (pH 4.1) and methanol, assisted by ultrasound. Then, supernatant was cleaned up by the solid phase extraction (SPE) with Strata-X cartridges and eluates were eluted with methanol at gravity flow. Once extracted, the analytes were identified by liquid chromatography-quadrupole time-of-flight mass spectrometry (LC-QqTOF-MS). As a result 50 compounds were identified, being the pharmaceuticals the main group of pollutants with 31 compounds. Human metabolites were the most relevant, present in all samples, including nucleotides (adenosine triphosphate), amino acids (phenylalanine) or drugs (icatibant, fenoprofen). On the other hand, several compounds were tentative identified and are pending of confirmation. The results of this study demonstrate the interest of high resolution mass spectrometry to draw the profile of contaminants in solid complex matrices. Furthermore, the data obtained provides information about the potential risk of use the sewage sludge for agriculture. Continue researching is needed to assess the real environmental risk related to this situation and the potential risk for human health. However, reports on CBs in aquatic organisms-especially the aquatic organisms in typical epidemic areas of schistosomiasis prevalence in China-are lacking. The release of CBs from the production and use of Sodium pentachlorophenate (Na-PCP) has been identified as one of the most important sources. Dongting Lake is the second largest fresh water lake of China, which is also an area with most widely distributed onchocerciasis and has the most severe schistosomiasis epidemic situation in China. Na-PCP has been sprayed as molluscicidal in Tianjin Dagu Chemical Company until 2003 with a production quantity of about 2000 tons/yr. CBs have been detected in water samples of Dongting Lake, but it was still produced as an intermediate of pentachlorophenol in Tianjin Dagu Chemical Company until 2003 with a production quantity of about 2000 tons/yr. CBs have been identified in environmental samples of Dongting Lake, including crustaceans, water samples, and sediments. The analysis of these samples showed the presence of chlorobenzenes, including chlorobenzene, dichlorobenzene, trichlorobenzene, and tetrachlorobenzene. The concentrations of these compounds were below the detection limit in all samples. However, the presence of CBs in environmental samples suggests that these compounds may have a potential risk to human health and the environment. The study of the fate and effects of CBs in aquatic ecosystems in Dongting Lake is important to understand the potential risks and to develop strategies to mitigate these risks.
Wastewater represents an alternative option for use in urban areas, industry and, especially, agriculture. Pharmaceuticals may undergo incomplete elimination in wastewater treatment plants (WWTPs) and are found in reclaimed wastewater, possibly being uptaken in crops following wastewater irrigation. Among commonly consumed crops, vegetables present considerable amounts of contaminants, possibly being removed by washing. In this study, we investigated the fate of pharmaceutical compounds in a wastewater treatment plant (WWTP). Model parameters were adapted to the situation at site. Chemical data were estimated using ACDCiLab. Model predictions were verified with measurements from a monitoring campaign in the WWTP. Results showed a high measured removal efficiency of Diclofenac, Ibuprofen and Ketoprofen concentrations in the WWTP and the simulation tool confirmed the same conclusion. As to uptake in lettuce, empirical results were compared to simulation outcome. For plant uptake prediction, a new steady-state model with translocation and phloem flow was applied [2]. Addition of phloem transport was necessary because the investigated compounds include weak acids (pKa 4-5), sugar, and phylloidal and phloem sap. Different methods for the quantitative analysis of OCPs was developed and validated by gas chromatography (GC). The method was established using the modified QuEChERS method for OCPs in orchard soil and grape, peach, apple, and pear. Recovery and limit of detection (LOD) of OCPs in orchard soils and fruits were 74.4-115.6 and 74.7-92.4%, 0.04-0.08 and 0.2-0.4 µg/g, respectively. The precision was reliable since the reproducibility of the extraction method (daily deviation) was less than 20%, which was the normal percent value. The residue of OCPs in orchard soils was analyzed by the developed method, and endosulfan sulfate, 2,4-DDT, 4,4-DDT, 4,4-DDD, and 4,4-DDE were detected at 1.1-444.9, 2.2-31.9, 4.5-863.1, 1.9-48.0, and 2.3-193.9 µg/kg, respectively. But OCPs in grape, peach, apple, and pear were not detected in all samples. These results showed that the residue in orchard soil were lower than bioaccumulation occurring.

PhytoCOTE project: Assessment of organic and inorganic contamination in vineyard soils M. Stutt, LPTC / EPOC UMRS 5805; E. Stutt, University of Danmark / DTU Environ; I. van Altena, University of Amsterdam / Plant Protection Institute; M. van Altena, University of Amsterdam / Plant Protection Institute. The project investigated the fate of pharmaceuticals in a wastewater treatment plant (WWTP), which included a system for the elimination of microplastics in wastewater sludge. The sludge was used as fertilizer in agriculture farming. This study focuses on the occurrence of pharmaceuticals in the size range 5000-10 µm in soils that received wastewater sludge as fertilizer. It presents the methods of sample preparation and presents field data. In Sweden 3 fields were sampled. Monitoring of microplastic in sludge fertilizer have been spread over a period of 35 years. The fields have either received 3 tons/year, 1 ton/year or no sludge fertilizer. 40 kg of soil were sampled from each field. The method is an effective and robust method that can be used to identify and quantify microplastics. The results will be presented from the analysis of priority chemicals in organic materials prior to spreading and the data will be used to undertake a refined risk assessment and to calculate a maximum safe spread rate for each material considered for application to land.

Microplastics in Agriculture Soil. K.B. Olesen, Aalborg University / Department of Civil Engineering; N. van Altena, University of Amsterdam / Plant Protection Institute; J. van Altena, University of Amsterdam / Plant Protection Institute. Microplastic is an environmental pollutant of worldwide concern. However, neither microplastic concentrations nor their sources or sinks are completely known. The project investigated the fate of pharmaceuticals in a wastewater treatment plant (WWTP), which included a system for the elimination of microplastics in wastewater sludge. The sludge was used as fertilizer in agriculture farming. This study focuses on the occurrence of pharmaceuticals in the size range 5000-10 µm in soils that received wastewater sludge as fertilizer. It presents the methods of sample preparation and presents field data. In Sweden 3 fields were sampled. Monitoring of microplastic in sludge fertilizer have been spread over a period of 35 years. The fields have either received 3 tons/year, 1 ton/year or no sludge fertilizer. 40 kg of soil were sampled from each field. The method is an effective and robust method that can be used to identify and quantify microplastics. The results will be presented from the analysis of priority chemicals in organic materials prior to spreading and the data will be used to undertake a refined risk assessment and to calculate a maximum safe spread rate for each material considered for application to land.
Pharmaceuticals uptake by spinach from seven soils mixed with sewage sludge

Two experiments were performed. a) Soils mixed with sludge were packed in SEM (Cambisol). Sewage sludge samples were taken from two wastewater treatment plants. b) Plants were exposed in the laboratory to different pollutants. The plants were exposed in the laboratory to different pollutants. Initially, the prevalent diclofenac was chosen. After the plants reached their maturity, they were exposed to pollutants for a few days. To establish the concept at the beginning comparably high concentrations of pollutants were applied. Finally, we will be able to provide an open access database of plant metabolites (PHARMITIDES-IDENT) and implement it into an analytical platform constructed earlier (FOR-IDENT; see https://water.for-ident.org). This work is supported by the Bavarian State Ministry of the Environment and Consumer Protection.

Pharmaceuticals uptake by spinach from seven soils mixed with sewage sludge

This study aimed to contribute to this growing area of research by exploring new or modified secondary metabolites that appear after addition of pollutants. To the best of the authors knowledge, no research is available up to now that surveyed the changes in the plant metabolite pathways of constructed wetland plants (CWP). Moreover, along with growth in CWP, due to possibly accumulated contaminants there is increasing concern about how those plants must be treated further, i.e., which points have to be considered regarding their disposal. We will provide a conceptual theoretical framework based on analysis of different plant extracts before and after exposure to different pollutants using novel RPLC-HILIC/ToF-MS technique. The plants were exposed in the laboratory to different pollutants. Initially, the prevalent diclofenac was chosen. After the plants reached their maturity, they were exposed to pollutants for a few days. To establish the concept at the beginning comparably high concentrations of pollutants were applied. Finally, we will be able to provide an open access database of plant metabolites (PHARMITIDES-IDENT) and implement it into an analytical platform constructed earlier (FOR-IDENT; see https://water.for-ident.org). This work is supported by the Bavarian State Ministry of the Environment and Consumer Protection.

Pharmaceuticals uptake by spinach from seven soils mixed with sewage sludge

This study was focused on a mobility of pharmaceuticals from sewage sludge in soils and their uptake by plants. Soil samples were taken from top horizons of seven different soil types (Stagnic Chernozem Silte, Haplic Chernozem, Gleyic Phaenozem, Haplic Lavisol, Arenosol Epieutric, Haplic Cambisol, Dystric Cambisol). Sewage sludge samples were taken from two wastewater treatment plants. Two experiments were performed. a) Soils mixed with sludge were packed in plastic columns, humidified to a value close to a field water capacity and 14 days incubated under laboratory conditions. Then, a ponded infiltration was applied and cumulative water outflow and solutes discharge from the bottom were measured. b) Spinach (Spinacea oleracea L.) was planted in soils mixed with sludge packed in plastic columns under greenhouse conditions. The amount of pharmaceuticals in plant parts (i.e., roots and leaves) was evaluated after harvesting. Compounds’ discharges as well as their root uptakes were soil and sludge dependent. In general, mostly larger discharges were observed for the Arenosol Epieutric and Cambisols. Mobility of compounds depended on their sorption affinity to particular soil types, i.e., water content or non-sorbed slurry can increase soil pH and urease activity, and non-sorbed sludge can promote soil laccase activity. The results show that the dissipation of DEHP is accelerated after incubation with SMS for 25 d, however little effect can be found with continued incubation due to low DEHP bioavailability. In this research, SMS amendment exhibits no effect on DnBP dissipation in soils and DnBP accumulation in bok choy. It was proposed that atmospheric deposition of DnBP to the site might be the main source of DnBP in bok choy in the study, since equivalent amounts of DnBP were detected in the vegetables grown in soils with or without DnBP spiking. This study indicates that the application of SMS as an organic fertilizer is less likely to affect the fate of PAEs in soils, and proper strategies should be conducted to reduce PAE levels in atmosphere to control PAE contamination in vegetables.

Plants: predicting and assessing direct, indirect effects and recovery of plants from chemical stress (P)

Experiences of demonstrating aquatic plant recovery following herbicide exposure using sloped mesocosms

F. Pickering, Cambridge Environmental Assessments

Recovery is important when considering short term exposure of aquatic non-target plants to herbicides in edge of field water bodies. Mesocosms studies allow the effects on both individual species and communities to be assessed simultaneously. Unlike indoor laboratory studies, where test item concentrations are artificially maintained, mesocosms studies allow for a more realistic application and dissipation of test item. Therefore, mesocosms studies can assess direct and indirect effects whilst a test item is present, and also monitor the period after dissipation has occurred to assess possible recovery. Using our sloped mesocosms at Cambridge Environmental Assessments (CEA), we are successfully able to test up to ten macrophyte species, each with different physical structures and characteristics (e.g. rooted; emergent). Some traditional measures of plant health can be seen as subjective (e.g. necrosis scoring and macrophyte mapping). At CEA, additional metric parameters such as number of nodes, number of leaves and stem lengths are routinely measured throughout the in-life phase. As a result, a combination of parameters are measured for each species tested. This ensures that the endpoints of a study are suitably robust and can be used to assess recovery. Here we will use results from our past studies to share experiences of assessing plant health. We will discuss which endpoints are most sensitive, reliable and therefore most suitable for determining effects on each plant species. Finally we will discuss which of these measurements are most relevant to assess recovery.

Impact of plant density on the end points (ER50) determined for crop protection products in Non Target Terrestrial Plants Studies conducted to OECD 227, Vegetative Vigour

E. Paterson, A. Thompson, Dow AgroSciences; G. Meregalli, Dow AgroSciences; E. Hooper, Dow AgroSciences; G. Karaiskou, Cambridge Environmental Assessments; K. Ralston, Dow AgroSciences; G. Meregalli, Dow AgroSciences; E. Hooper, Dow AgroSciences; G. Karaiskou, Cambridge Environmental Assessments; K. Ralston, Dow AgroSciences

Non Target Terrestrial Plant (NTPP) studies conducted to OECD 227 test guidelines are submitted as part of the registration process for plant protection products in Europe and the US. Current planting densities described in OECD test guideline 227 for Vegetative Vigour Studies, recommends 1-2 large plants per 15 cm and 1-3 medium size species per 15 cm pot and 5-10 small species per 15 cm pot. The minimum recommended number of plants per treatment in OECD 227 is 20, as a result vegetative vigour studies conducted to this guideline, regularly exceed 750 pots. These studies require a large amount of glasshouse space and are very labour extensive. Establishing, spraying, assessing and handling data from such large studies has its own issues and can increase the probability of errors occurring during the study, which can be costly and time consuming to the researcher. Unlike indoor laboratory studies, where test item concentrations are artificially maintained, mesocosms studies allow for a more realistic application and dissipation of test item. Therefore, mesocosms studies can assess direct and indirect effects whilst a test item is present, and also monitor the period after dissipation has occurred to assess possible recovery. Using our sloped mesocosms at Cambridge Environmental Assessments (CEA), we are successfully able to test up to ten macrophyte species, each with different physical structures and characteristics (e.g. rooted; emergent). Some traditional measures of plant health can be seen as subjective (e.g. necrosis scoring and macrophyte mapping). At CEA, additional metric parameters such as number of nodes, number of leaves and stem lengths are routinely measured throughout the in-life phase. As a result, a combination of parameters are measured for each species tested. This ensures that the endpoints of a study are suitably robust and can be used to assess recovery. Here we will use results from our past studies to share experiences of assessing plant health. We will discuss which endpoints are most sensitive, reliable and therefore most suitable for determining effects on each plant species. Finally we will discuss which of these measurements are most relevant to assess recovery.

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WE154
Interspecific competition impact on organism responses to chemical stress: an SSD-based approach.
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Organisms are not alone in the environment. They interact with other individuals of the same or other species in different ways. Interspecific competition is an important interaction for herbaceous plants in grass strips. Such vegetated areas generally act as buffer strips against pollutant flows and are thus submitted to various chemical exposures. However, competition is rarely considered in environmental risk assessment. To address this point, we tested whether competition modifies the way plants respond to herbicide (isoproturon) toxicity in an attempt to link individual tolerance of organisms and community dynamic. Then we investigated the impact of competition on species sensitivity distribution (SSD), a widely used community-level risk assessment tool that usually considers monospecific bioassays only. To do so, we exposed during 25 days 6 herbaceous species (representing various isoproturon tolerance and competition ability) to 6 isoproturon concentrations (0 to 1.75μM) in presence and absence of a selected competitor, Bromus erectus (choice based on its high resistance to isoproturon and its high competition ability). For each setting, we replicated 5 replicate treatments, e.g. by degree exposure or transport. In such cases, the plants might recover if the effects are reversible. Neglecting the recovery potential in the risk assessment is definitely protective but might be over-restrictive in the case of unnecessary losses of crop yields. The recovery subgroup of the SETAC Plants Interest Group aims to review the different approaches to analyse recovery of plants and to make suggestions how recovery of these traits can be included in a risk assessment framework. In the present contribution, we will focus on aquatic algae and macrophytes and the regulation of plant protection products in the EU. Experimentally, recovery of algae and macrophytes can be assessed in single species laboratory tests or micro- and mesocosm studies. In refined exposure laboratory toxicity tests, usually the recovery of the growth rate is assessed since the populations are kept in the experimental growth phase. In micro- and mesocosm studies, it is possible to analyse also recovery of biomass or biomass and potential indirect effects. The differences of these two options and their potential consequences for risk assessment will be discussed. Effect modelling can be used to extrapolate from empirical data to other exposure scenarios or species. However, while the simulation of refined exposure laboratory toxicity tests seems to be straightforward, the prediction of effects under field conditions is still challenging. In addition, the use of such models in the risk assessment requires clearer criteria on which magnitude and duration of effects can be considered acceptable.

WE156
Rimsulfuron toxicity and recovery in duckweed (Lemna minor) M. OPINCARNE, University of Florida / School of Natural Resources and Environment; P.C. Wilson, Z. Li, University of Florida IFAS / Soil and Water Science
Rimsulfuron is an herbicide for which very little is known about its toxicity to aquatic macrophytes. This study was designed to evaluate the effects of rimsulfuron on the model aquatic macrophyte Lemna minor at low concentrations. This study also evaluated recovery by L. minor following a 5 day exposure period. Growth rates were measured at 1, 3, and 5 days following exposure to rimsulfuron-fortified 10% Hoaglands media at concentrations of 0, 0.0003, 0.0006, 0.00125, 0.0025, 0.005, 0.01, and 0.02 mg/L. After 5 days exposure, growth rates were significantly lower for rimsulfuron concentrations ≥0.0006 mg/L. Following the 5-day exposure period, plants were rinsed with deionized water and placed in fresh Hoagland’s nutrient media. Impact on growth rate was measured as percent impact compared with growth of the control. Growth rates for 00006 mg/L were reduced 25.4% relative to the controls. Interestingly, a horometic response was observed at the 0.0003 mg/L concentration. In this case, the growth rate was 16.7% relative to the control. Following exposure, signs of reduction in growth rate were observed on days 3, 5, and 10. However, 15 days after removal from the rimsulfuron treatment solutions, growth rates recovered to control levels. While rimsulfuron exposure significantly reduced growth rates of L. minor at all concentrations ≥0.0006 mg/L, effects were found to be reversible. Rimsulfuron was fast acting, with toxicity observed 1 day following exposure. In contrast, a longer period of time was required for growth rates to recover to control levels.

WE157
Toxicokinetic/toxidynamic (TK/TD) modelling - Increasing the realism in risk assessments for aquatic plants S. KIM, Bayer AG / Effect modelling; E. Bruns, Bayer AG, Division Bayer CropScience / Ecotoxicology; A. Solga, Bayer AG; T. Preuss, Bayer AG / Environmental Safety
For assessing the risk of plant protection products (PPPs) to aquatic ecosystems, environmental concentrations of the active substance need to be estimated. Throughout Europe different approaches are used to predict these environmental concentrations. To characterize the in-field performance of a herbicide (e.g. rimsulfuron, Rims) as aquatic plants, ecotoxicological thresholds derived from experiments with constant exposure to an active substance over several days are compared to the predicted maximum environmental concentration. Although, it can be deemed conservative to only consider the maximum concentration, there are plenty of cases where the risk assessment becomes overly conservative due to this practice. This applies particularly to assessments for lots, bodies (streams) in which critical peak concentrations usually last for a few hours or days. Since some models for the prediction of environmental concentrations do not only deliver maximum concentrations but also temporally explicit exposure (exposure patterns), a more detailed and realistic assessment of exposure is possible. To also increase the realism on the effect side, either an ecotoxicological threshold from a refined exposure experiment is needed, or the effect of the predicted exposure pattern on the organism is investigated by ecological modelling. We propose TK/ TD modelling as a powerful tool to evaluate effects of time variable exposure on aquatic plants. TK/ TD modelling refers to linking effects to the internal concentrations in an organism instead of the external one and by this being able to consider time-variable exposure patterns. For characterizing risks of active substance exposure by TK/ TD modelling, it is necessary to adjust the approach to a specific substance. Adjusting in this context means defining TK/ TD parameters to describe the uptake/elimination and the internal dose-response relationship. Besides defining the parameters, it is also necessary to validate them by using the parameterized TK/ TD model and by comparing predictions of the model to measured data. In this work, we report on the development of a Lemna minor TK/ TD model that was parameterized to describe the effects of different sulfonyleurea herbicides. The results demonstrate that the TK/ TD Lemna model with its specific parameterization is able to reliably predict effects. Using the TK/ TD Lemna model allows to perform a more realistic environmental risk assessment and to link time variable-exposure to effects.

WE158
Assessing soil toxicity of methylparaben using plants and collembola D. Kim, J. Kwak, Konkuk University; Y. An, Konkuk University / Department of Environmental Health Science
Methylparaben is an endocrine disrupting chemicals (EDCs) and is contained in personal care products such as cosmetics and quasi-drugs. Methylparaben is known to have low toxicity to mammals, but there is no data on hazard assessment for soil ecosystem. Methylparaben was mostly removed in the sewage treatment process, but was detected in soils of various countries. In addition, there is a possibility that personal care products may leak into aquatic or soil environments if they are not disposed of properly. To overcome this, it is necessary to evaluate the hazard assessment of methylparaben in soil ecosystem. This study assessed the toxicity of methylparaben to plants (mung bean and rice) and collembola. Plants were exposed methylparaben from 0 to 400 mg/kg for 14 and 21days. In plant toxicity tests, shoot and root growth, root development, stomatal opening size, chlorophyll contents and photosynthetic factors were measured. In the collembola test, methylparaben was exposed at 0 to 500 mg/kg for 5 days and mortality was observed. The most sensitive endpoint in mung bean was identified as stomatal opening size, and no observed effect concentration (NOEC) was 10 mg/kg. The most sensitive factor in rice was chlorophyll contents, and NOEC was under 10 mg/kg. The half-lethal concentration (LC50) value for collembola was 440.53 mg/kg. Methylparaben appears to have significant physiological effects on plants even at low concentrations. The results of this study can be fundamental for soil risk assessments of methylparaben. This study was funded by the Korea Ministry of Environment (MOE) as the Environmental Health Action Program (1485014458), and the Graduate School of Specialization for
WE159 Evaluation of phytoxicity for Bisphenol A with new endpoint, phytoestrogen D. Kim, J. Kwak, Konkuk University; Y. An, Konkuk University / Department of Environmental Health Science

We demonstrated that endocrine disrupting chemicals (EDCs) are known as chemicals that show hormone-like action or inhibit hormones, the phytoxicity assessment of EDCs does not have any specific toxic endpoints for these substances. The factors (growth, photosynthetic activity, chlorophyll, etc.) used to evaluate common toxic substances such as heavy metals are also applied to EDCs. These factors are not suitable for EDC materials, which have relatively low toxicity to organisms, and phytoestrogen. However, if associated with phytoestrogen, takes a long time to assess toxicity. Therefore, we tried to evaluate phytoestrogen, a new toxic endpoint for EDC materials, using bisphenol A. Meanwhile, bisphenol A is known as a representative EDCs used in the production of consumer products and in various industrial fields. While it is used widely for various purposes, the soil ecotoxicity of bisphenol A is limited. Therefore, we evaluated the toxicity of bisphenol A on Paracllorella kessleri, an alga (green algae) used for traditional endpoints and evaluated the applicability of phytoestrogen, a new endpoint for EDCs materials. This study was funded by the Korea Ministry of Environment (MOE) as the Environmental Health Action Program (1485014458), and funded by the Graduate School of Specialization for managing information related to chemical risk.

WE160 Soil toxicity of DEHP and Nonylphenol on mungbean and rice D. Kim, J. Kwak, Konkuk University; Y. An, Konkuk University / Department of Environmental Health Science

DEHP is used as a plasticizer and insecticide. Especially, it can be used as plastic vinyl applied in farmland. Nonylphenol is used as surfactant, and 4-nonylphenol is used as a synthetic substance with various isoforms which are used as insecticide and detergent. Although DEHP and nonylphenol are likely to release into the soil environments, soil ecotoxicity data are currently limited. Ecotoxicological researches in soil were reported in only three cases of DEHP and four cases of nonylphenol. This study was conducted to evaluate the effects of DEHP and nonylphenol on the growth and physiological changes of mung bean and rice. The toxicity tests were conducted on 14 days (acute) and 21 days (chronic). Shoot growth was measured in 14 days-acute experiment and physiological factors including stomata opening size, chlorophyll contents, and photosynthetic activity were evaluated in the 21 day-chronic experiment. This study is meaningful because the soil toxicity of the two substances to the plants was conducted using various factors, and the results of this study can be fundamental for soil risk assessments of DEHP and nonylphenol. This study was funded by the Korea Ministry of Environment (MOE) as the Environmental Health Action Program (1485014458), and the Graduate School of Specialization for managing information related to chemical risk.

WE161 Toxicity of a glycosylated based formulation on phytoplanktonic green microalgae J.G. Perez, Universidad de Buenos Aires, Facultad de Ciencias Exactas y Naturales / Departamento de Biodiversidad y Biología Experimental, Buenos Aires; A. Magdaleno, Universidad de Buenos Aires / Facultad de Farmacia y Biociencias, Cátedra de Salud Pública e Higiene Ambiental; M. D. Rivero de Molina, Universidad de Buenos Aires, Facultad de Ciencias Exactas y Naturales / Departamento de Química Biológica, CONICET-UBA, Instituto de Química Biológica - Ciencias Exactas y Naturales (IQUBICEN); A.B. Juarez, Universidad de Buenos Aires / Facultad de Ciencias Exactas y Naturales, Departamento de Biodiversidad y Biología Experimental

The emergence of transgenic crops led to an increase in the use of glyphosate and its presence in different ecosystems is a worldwide problem. Although it was designed to inhibit the aromatic amino acids synthesis in plants, glyphosate exerts toxic effects on non-target organisms, probably through other mechanisms. Its entry into waterbodies means water is a risk for biota, particularly for the phytoplankton microalgae community and the results of this study can be fundamental for soil risk assessments of glyphosate and nonylphenol. This study was funded by the Korea Ministry of Environment (MOE) as the Environmental Health Action Program (1485014458), and the Graduate School of Specialization for managing information related to chemical risk.

WE162 Indicator, indigenous and invasive species: the need of risk-benefit considerations in PPP risk assessment G. Meregalli, Dow AgroSciences Italia s.r.l. / Ecotoxicology; C. Vaj, V. Zaffagnini, A. Carone, Dow AgroSciences Italia srl

Indicator species are the basis of the ecological risk assessment framework. Endpoints derived for these species are used in the risk assessment to evaluate the safety of, e.g., plant protection products (PPP) to non-target species. In certain cases indicator species are also indigenous species to a particular region (e.g. Myriophyllum spicatum in Europe). Invasive species are non-autochthonous species, accidentally introduced in a given region, which, in absence of their natural predators, often grow uncontrolled and overcome indigenous species, completely devastating the biodiversity of the habitats they colonise. The uncontrolled growth of these species can be also a threat to ecosystem functioning, e.g. altering oxygen balance in the case of the aquatic environment or shifting the prey/predator equilibrium. In cases where other control means are not possible, PPP could be employed to control the spread of invasive species. As an example, in the USA some herbicides have been authorised to control M. spicatum, which is an invasive alien species in North America. On the other hand, recently in Europe there have been reports of Myriophyllum aquaticum, a new alien invasive species genetically related to the indicator M. spicatum. In Piedmont (Italy), M. aquaticum has been observed in the Po River, threatening aquatic biodiversity. In addition also rice cultivation, a very important crop for the region, is at risk, due to the uncontrolled growth of M. aquaticum in canals and ditches feeding water to rice paddies. This species has been added as an invasive species to the black lists of Piedmont and of the European Union and it is being monitored and controlled with local initiatives. To prevent the irreversible degradation or destruction of natural habitats by non-autochthonous species, a timely intervention may be necessary, even if this may result in a temporal impact on indigenous desirable species. Programs to prevent the invasion of species are being developed to reduce the impact of alien species on rice ecosystems. The use of a risk-benefit analysis, which can help in the decision of whether PPP are suitable for use in such programs, will be discussed in light of the two abovementioned examples.

WE163 Auxinic herbicides: the impact of water plants’ root measurements on the risk assessment G. Gonsior, Eurofins Agroscience Services Ecotox GmbH

Data requirements for the registration of plant protection products in the EU Regulation 1107/2009 indicate that a test on a Myriophyllum species is necessary for auxinic herbicides. The OECD 239 water sediment test with Myriophyllum spicatum species developed in North America. The other hand, recently in Europe there have been reports of Myriophyllum aquaticum, a new alien invasive species genetically related to the indicator M. spicatum. Programmes to prevent the invasion of species are being developed to reduce the impact of alien species on rice ecosystems. The use of a risk-benefit analysis, which can help in the decision of whether PPP are suitable for use in such programs, will be discussed in light of the two abovementioned examples.

WE164 Testing the emergent macrophyte, Glyceria maxima in a water-sediment system: Results of a ring-test with Isoproturon J. Davies, Syngenta / Environmental Safety; G. Arts, Wageningen Environmental Research (Alterra) / Environmental Risk Assessment; K. Kuhl, Bayer AG - Crop Science Division; J. Kubritz, BASF. M. Ratte, ToxRat Solutions GmbH & Co. KG

Under EU pesticide regulation, regulatory tests are required for the aquatic macrophyte, Lemma, and two algal species for herbicides and plant growth regulators. Data requirements introduced under EU Directive 1107/2009 stipulate that further tests may be required for compounds which show selectively higher toxicity to either dicotyledonous or monocotyledonous plant species in terrestrial plant tests. In these cases, the recommended dicot and monocot species
are Myriophyllum and Glyceria, respectively. OECD Test Guideline 239 for testing Myriophyllum spicatum in a water-sediment system was adopted in September 2014 and this test method has since been adapted to facilitate growth of the emergent, reed grass, Glyceria maxima. During 2016 and 2017, 15 laboratories participated in a ring-test with the herbicide, isoproturon against Glyceria. The objectives of this test were to establish standard incubation techniques, to determine the required test concentration, to characterize control variability and inform test design and to identify appropriate validity criteria. Results of this ring-test will be presented alongside progress on a second ring-test with the herbicide imazaquin, scheduled for Spring / Summer 2018.

**WE165**
**Study of the toxicity effects of Cd, Ni and Zn on macrophytes, antioxidant responses and time for steady-state bioaccumulation under constant metal concentrations exposures**

S. martínez, CONICET PRIET UNEU; M. Saenz, PRIET CONICET, National University of Luján; W.D. Di Marzio, CONICET-PRIET / PRIET

Heavy metals represent an actual environmental problem because the industrial and commercial uses of them are continuously increasing, bringing on a widespread contamination. Examples of human activities that contribute in heavy metal contamination are mining, smelting, fossil fuel combustion and industrial applications. Human activities may also indirectly cause changes in the environment that mobilize metals that were otherwise bound in stable forms, making them bioavailable. Aquatic bodies directly or indirectly receive pollutant discharges and metals faciity in water are easily absorbed by organisms. This study was aimed to evaluate the individual toxicity effect of Cd, Ni and Zn on a macrophyte and its bioaccumulation capability. Cadmium is a hazardous environmental pollutant and is toxic to most organisms. Nickel and Zinc are essential trace elements needed in the nutrition of plants. Nevertheless, over certain threshold they can present phytotoxic activity. Macrophytes are key elements in aquatic ecosystems and are used for treatment of contaminated water areas. Ceratophyllum demersum. Following the OECD 2014 guideline for sediment-free toxicity test, plants were exposed to a range of concentrations (1-16 Ni, 4-64 Zn or 0.5-8 Cd mg/L) and fresh weight, main shoot length and total shoot length were chosen as endpoints. For the bioaccumulation assays, plants were exposed to a constant metal concentration throughout the period of time until internal metal concentration reached a steady state. To make sure of a constant external metal concentration, a daily renewal of the media was carried out. Besides, the influence of these metals on antioxidative enzymes activity was evaluated for the three lower concentrations of each one. These enzymes are involved in the plant defense mechanisms activated by heavy metal exposure. Determination of catalase (CAT), glutathione peroxidase (GPOX), ascorbate peroxidase (APOX) was performed from the total plant mass. In the main, fresh weight resulted in the most sensible endpoint reaching an inhibition of almost 50% for 64 mg Zn/L and presenting a dose response. A dose response was observed during plant growth using UHPLC/DAD/MS in SETAC Europe 28th Annual Meeting Abstract Book

**WE166**
**Physiological responses of Thlaspi prae cox (Brassicaceae) to Ni hyperaccumulation**

T.D. Mišljenović, K. Jakovljević, S. Jovanović, University of Belgrade / Faculty of Biology Institute of Botany and Botanical Garden Jevremovac; N. Mihailović, University of Belgrade / Institute for the Application of Nuclear Energy; V. Maksimović, University of Belgrade / Institute for Multidisciplinary Research; D. Mišić, University of Belgrade / Institute for Biological Research Siniša Stanković Thlaspi prae cox is a well known heavy metal hyperaccumulating plant species. The ability of T. prae cox to hyperaccumulate Zn and Cd have been extensively studied, while data on Ni hyperaccumulation are scarce. Our aim was to bring more understanding to the physiology of T. prae cox exposed to increasing concentrations of Ni. Seeds of T. prae cox were collected from an ultramafic site on Mt. Maljen (Serbia). Two - week old seedlings were transplanted to a peat based substrate amended with increasing concentrations of Ni (250, 500, 1000 ppm). Plants were grown for 3 months under controlled conditions. Content of Ni in plants was analyzed by ICP, while phenolics, sugars and organic acids have been analysed using UHPLC/DAD/MSS or HPLC-PAD. No visible toxicity symptoms were observed during plant growth, and Ni did not affect biomass production at applied concentrations. A dose-dependent response of T. prae cox shoots to applied Ni concentrations was recorded. Ni hyperaccumulation threshold of 1000 ppm was reached in the plant. The harvested plants were dried in vacuum oven and a microwave acid digestion were carried out. Contents of phenolic acids and flavonoids were generally low, and were not significantly affected by increasing Ni concentrations. Effects of increasing Ni concentrations on the content of sugars and organic acids in shoots have also been analysed.

Understanding the physiology of T. prae cox exposed to Ni and its Ni tolerance limits might be relevant for the potential application of this species in phytostabilization or phytoextraction technologies at contaminated soils.

**WE167**
**Phytoextraction of heavy metals in Cienega of Tamasopo wetland, México, by Typha latifolia**

C. Wong, C. Carranza, Universidad Autonoma de San Luis Potosi / Laboratorio de ecotoxicología; A.J. Alonso, Universidad de Guanajuato / Departamento de Farmacia

Heavy metals are persistent inorganic toxic pollutants that come from diverse anthropogenic activities [1]. They can be easily absorbed by the roots of plants due to their relatively high mobility in the soil. The mobility of many heavy metals is known to have great importance, forming a substantial component of the primary production in many aquatic ecosystems, especially in wetlands. Plants can remove and accumulate metals from the solution by phytoextraction; however, the metals can also be precipitated or eliminated from the solution by ion exchange or by adsorption on organic and inorganic compounds. Concentrations of heavy metals in aquatic plants depend both on metal speciation and on the species of plant absorbing the metal [2]. High concentrations of some trace metals in aquatic plants have led many authors to believe that they accumulate from water and/or from sediments; the uptake is influenced by several factors, such as temperature, pH, light and the presence of other metals in the water, all of which alter the uptake of heavy metals into the tissue [3]. Metal uptake by plants has three patterns: (1) true exclusion in which metal is not removed from entering the plants; (2) shoot exclusion, in which metals are accumulated in the root but translocation to the shoot is restricted; and (3) accumulation, where metals are concentrated in the plant parts [4]. The present research examines the phytoextraction in situ of heavy metals by Typha latifolia to determine the concentration of these metals in the plant, water and sediments. The experimental procedure consisted of: 1) sampling of five sites of the Cienega de Tamasopo wetland; the sites were characterized in terms of their C. Wong, C. Carranza, Universidad Autonoma de San Luis Potosi / Laboratorio of Typha latifolia. 3 samples of 5% acidified water with HNO3, and one sample without acidification for physicochemical parameters, one sample of the 1st cm of sediment; 2) plants: washing, separation in roots and leaves and drying at 70 °C for 18 hours in Lindberg / Blue stone; 3) grading and spraying of root and leaves in analytical mill (KIKI Werke M20); 4) acid digestion with HNO3/3HCl in plate at room temperature; 5) quantification of heavy metals by ICP-MS in digestion and water and column samples. The results show that Typha latifolia accumulate Mn>Zn>Cr>Pb>Cu>As>Hg>Cd in roots. This study aimed to gain a better understanding of the importance of aquatic plants such as Typha latifolia in heavy metal accumulation and detoxification mechanisms.

**WE168**
**Heavy metal removal by aquatic plants**

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Removal of heavy metals from the environment due to industrialization and urbanization is a great problem worldwide, due to their toxicity to many life forms. Aqueous waste from metal plating, mining operations, tanneries, smelting, alloy industries, and storage batteries are sources of metal contamination. Biological methods have been recommended as effective alternative for metal recovery of heavy metals from aqueous solutions. Aquatic plant biomass represents an important biological source for the removal and accumulation of heavy metals and therefore have been exploited worldwide in the field of wastewater treatment technologies. Aquatic plant species including free floating and submerged, as Lemna, Spirodella, Ceratophyllum and Myriophyllum, have shown potential for metal removal from wastewater. The aim of this study is to evaluate the efficiency and capacity of different species of aquatic macrophyte in the removal of heavy metals from the solution in a multi metal solution. The initial whole ecotoxicity of a multi-metal system composed by Cd, Ni and Zn was assessed by growth inhibition test with the green alga P. subcapitata, acute toxicity test with D. magna and ex vivo cytotoxicity test with E. foetida coelomocytes. An experiment was set up for 10 days, by the addition of 10 grs of fresh weight of plants from different species mentioned above, in the metals solution. Previous works have shown that metal uptake rates were faster within the first 48 hours, and decrease with time and with metal concentration solution, so, at this time and at 3 and 5 days, respectively, plants were removed and new plants were placed in the same multi-metal system. At these time intervals, samples of solution and plant were taken for metal determination. The harvested plants were dried in an oven and a microwave acid digestion were carried out. Metal determinations in aqueous and plants sample were made by flame atomic absorption spectrometry. There was a gradual decrease in metal contents in the artificial wastewater at time intervals. The metal removal capacity was different for each species and for each metal. At the end of the experiment, the multi-metal system treated with aquatic plants was assessed with the same battery of tests used in the beginning. The whole ecotoxicity of the artificial wastewater decrease after treatment with aquatic plants, demonstrating an efficacy for metal removal and recovery.
WE169  Toxicity of the binary mixture Cd-Zn on Lemna gibba evaluated using morphological and oxidative stress enzyme endpoints
s. martinez, CONICET PRIET UELU; W.D. Di Marzo, CONICET-PRIET / PRIET; M. Saenz, PRIET CONICET, National University of Luán
The presence of metals in the environment represents one of the major concerns as they are persistent in nature, non-biodegradable and can bioaccumulate in living organisms and plants. Metals in aquatic ecosystems may have toxic effects on the trophic level composed partly by aquatic vascular plants, also called macrophytes. These organisms play a critical role in this environment. As a representative species of macrophytes, we worked with a rooted free-floating Lemna gibba. The metals evaluated here were Cd and Zn, individually and in mixtures. Exposures of plants would not be carried out in presence or absence of Cd and Zn for 7 days. Different endpoints were determined at the end of the assays. Number of fronds, fresh weight, fronds/colonies ratio, frond area and exes’ length are the determined morphological endpoints. Physiological changes were evaluated as enzymatic activity of catalase, ascorbate peroxidase and guaiacol peroxidase, determined at the lowest concentrations. Both metal concentrations, bringing about a 50% inhibition of frond number (EC50) was determined. In order to compare the sensitivities of the different endpoints, NOEC and LOEC toxicity indexes were calculated. For Cd, fresh weight and fronds/colonies ratio resulted in the most sensitive, while for Zn total area was the most sensitive. Even though there was no significant difference for guaiacol peroxidase activity for Cd, it presented an increase compared to control. While the other enzymes had activity levels similar to the control. In the case of Zn, catalase and ascorbate peroxidase presented increases. Where higher than Cd however both of presented significant differences with it. For the mixture analysis, multiple regression was used to fit the observed %frond number inhibition (%FNI) to dissolved metal concentration (M(Ma)). The negative value of the parameter of the interaction between Cd and Zn indicates alleviation of %FNI and toxicity. The concentration addition approach was evaluated by calculating the sum of effective concentrations on single EC50s (ΣEC50) for each mixture test case and the percentage ΣEC50 of all test cases resulted 1.13 suggesting that this mixture presents an additive toxicity to Lemna gibba. Enzyme activity was also calculated at the lower concentrations of the mixtures. In general an increase in the enzymatic activity was observed. Ascorbate peroxidase and guaiacol peroxidase presented the maximum increase, while catalase had a moderated activity rise.

WE170  Increase of tolerance of green algae as a tool in metal bioremediation
M. Saenz, PRIET CONICET, National University of Luán; F. Cassani, S. Martinez, s. curieses, J. Alberdi, CONICET PRIET UNELU; W.D. Di Marzo, CONICET-PRIET / PRIET
Presence of various metals in aqueous streams arising from the discharge of untreated metal-containing effluents into water bodies, is one of the most important environmental issue, as human health risks and harmful effect to living organisms occur. In the last decades the amount of Chromium in aquatic and terrestrial ecosystems has increased as a result of different human activities such as mining, chromite exploitation and associated period processes that remove the metals. In this study, we aimed to evaluate the use of preadapted strains to subletal concentrations of Chromium, into bioremediation of Chromium containing wastewater. Preliminary results will be shown related to the assessment of the potential of this strategy to increase tolerance of selected species in order to become an interesting tool in the field of bioremediation processes mediated by green algae. Two green algae species were used in this study: Chlorella pyrenoidosa and Nannochloris oculata. These species differ in its morphological structure and organization level as the former has a cenobial feature while the second a free unicellular one. Both strains were maintained by a year under subletal concentrations of Chromium, into bioremediation of Chromium containing wastewater. Preliminary results will be shown related to the assessment of the potential of this strategy to increase tolerance of selected species in order to become an interesting tool in the field of bioremediation processes mediated by green algae. Two green algae species were used in this study: Chlorella pyrenoidosa and Nannochloris oculata. These species differ in its morphological structure and organization level as the former has a cenobial feature while the second a free unicellular one. Both strains were maintained by a year under subletal concentrations of Chromium ranging from 0.42 to 73 mg/l. These concentrations were chosen base on previous experiments through range finding tests. Subtale solutions were renewed monthly and algal cells were subcultured in new medium. After the preadapted period, each sublateral exposed algal population from both strain and one which was never exposed to the metal, considered as the control, were centrifuged. An inoculum of know cell density was prepared with each pellet, and the algae were exposed to a wide range of Chromium concentration solutions. Samples of solution and algal cells were taken for metal determination in order to elucidate the mechanism of resistance origin. The harvested cells were centrifuged and a microwave acid digestion were carried out. Metal determinations in subletal solutions and in algal sample were made by flame atomic absorption spectrometry. Chromium accumulation and compartmentalization in algal cells would explained the increase resistance observed. Further studies relate to detoxification mechanisms and chelating internal molecules as phytochelatin will be conducted to unravel the tolerance mechanisms involved.

WE171  Ecotoxicological assessment of the iron mining waste from Mariana (Brazil) on terrestrial flora using different plant species
O.R. Alves, University of São Paulo USP / Department of Hydraulic and Sanitation; O.A. Bandeira, T.J. Pinto, L.P. Figueiredo, University of São Paulo USP; E. Espindola, University of São Paulo USP / Hydraulics and Sanitation
In Brazil it is very common to have mining waste placed in dams, especially in the Minas Gerais state. The deposit of mining wastes implies in risk of dam rupture, between other problems. In November 2015, the rupture of the “Fundão” Dam in the city of Mariana in Minas Gerais state was one of the most welfare disasters in Brazil. The rupture caused severe impacts to the terrestrial and aquatic environments, where tons of the waste has been placed. It is now matter of concern to study the effects of the mining waste deposition in soil to terrestrial flora in order to understand the real consequences to the environment and so be able to propose actions for restoration and management of the affected area. The main goal of this study was to evaluate the ecotoxicity of the mining waste that outpoured the Fundão Dam to ten different plant species (Avena strigosa, Pennisetum glaucum, Croatalaria juncea, Canavalia ensiformis, Cajanus cajan, Cajanus cajan, Dolichos lablab, Mucuna pruriens gray, Mucuna pruriens black and Lupinus albus). The ecotoxicological assays were performed using the mixture of a natural soil (NS) from Mariana (uncontaminated) and the waste (W). The combinations of mixture were: P1: 75% NS and 25% W; P2: 50% NS and 50% W; P3: 25% of NT and 75% of the W and P4: 100% W. The parameters evaluated were: Fresh and dry biomass (shoot and root), height, length of the longest root and seed emergence. All species, except Lupinus albus and Avena strigosa, had EC50 and/or EC10 in at least one of the seven parameters evaluated. The species that presented 50% inhibition of root growth were C. juncea (73.07%), P. glaucum (82.68%) and C. cajan (79.54%). The height of the first two species was affected by 20% in the proportions 76.81 and 85.91%, respectively. The proportions 87.32 and 40.61% of waste affected 50% of the length of the longest root of C. juncea and P. glaucum. The results showed that: the species tested presented different indices to soil fertility by mining waste; the variety of microorganisms of the Dinis mutants caused phytotoxic effects in all tested species; the most sensitive and least sensitive parameters, respectively, were root growth (root length and dry biomass) and seed emergence.

WE172  Mitigation of CuO nanoparticles microbial ecotoxicity by plant in an agricultural soil: plant variety matters
J.M. Martins, IGE UMR 5001 / Université Grenoble-Alpes; A. Cantarel, Université Claude Bernard Lyon1 / UMR Ecobiologie Microbiénne 5557; J. Gervaix, Université Claude Bernard Lyon 1 / UMR Ecologie Microbiénne 5557; A. Richaume, Université Claude Bernard Lyon 1 / UMR Ecologie Microbiénne 5557
New types of pesticides based on nanoparticles (NPs) are now being used to optimize phytosanitary treatments. However, they can generate soil contamination by metal-oxide NPs such as CuO-NPs which NPs toxicity is still largely unknown. Several studies showed the deleterious effects of metal nanoparticles (NPs) on soil microbial communities and reported the importance of soil microbial ecototoxicity of nanoparticles (NPs) in plant growth and the effects of nanoparticles on microorganisms and their metabolisms. However, it is not fully understood whether the plant modifies the microbial ecotoxicity of NPs because of organic matter enrichment in the rhizosphere through the root exudation and whether the plant variety mitigates the ecotoxicity according to plant traits. Endpoints that relate to soil fertility by mining waste; the variable microbial activity of the Dinis mutants caused phytotoxic effects in all tested species; the most sensitive and least sensitive parameters, respectively, were root growth (root length and dry biomass) and seed emergence.

WE173  Use of Posidonia oceanica as a potential bioindicator species of metal pollutants: cellular and molecular responses to mercury exposure
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The marine phanerogam Posidonia oceanica, thanks to its worldwide distribution 366

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and to its capability to accumulate certain environmental metal pollutants, would be a potentially valuable bioindicator species of metal pollutants. Mercury represents one of the most abundant marine pollutants in the Mediterranean Sea. In this study sublethal effects of this metal were investigated in P. oceanica. Several foliar shoots of this aquatic plant were treated for 4 days (96 h) with different mercury concentrations (0, 0.1 and 1 μg L−1 Hg Cl₂) under constant laboratory conditions. Biochemical markers of oxidative stress of aquatic plants, such as the glutathione S-transferase activity, the ascorbate peroxidase activity, the total antioxidant capacity, the phenols content, the level of lipid peroxidation and the mononucleotides frequency were measured in different parts of adult leaves: the blades and the sheaths for antioxidant responses, the meristem and the sheaths for genotoxic effects. Although a limited effect of Hg was measured in analyzed tissues, a significant difference in localization of responses was found in leaves of all treatments.

WE174

Influence of toluene vapor exposure on plant metabolic changes

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The conventional damage diagnosis methodology for plants after chemical accident only relied on the change in their phenotype such as leaf-browning, so there had been lots of controversy because of uncertain causality and inaccuracy. The ministry of environment tried to characterize of plant damage by introducing metabolicomics using GC/MS and LC/MS as a powerful tool for sensitive damage diagnosis. However, target metabolite selection process was unclear and the exposure method did not reflect the chemical accident scenario, so the research results have not been put to practical use. Therefore, untargeted metabolomics and vapor exposure chamber were introduced in this study to overcome the limitations of existing research. The development potential of metabolomics-based damage diagnosis tool was demonstrated using Ulmus minor, Epicarpaea thyrsoides, and Prunus africana. Toluene was selected as target compound based on the scoring system, which takes into account both accident frequency and hazards. To reflect the realistic chemical accident scenario, plants were exposed in vapor exposure chamber. In this study, the metabolomics responses of plants at early development stages (4th leaf stage) to toluene were evaluated by liquid chromatography quadrupole time-of-flight mass spectrometry (LC-QToF-MS) based untargeted metabolic profiling. The exposed concentration-based and recovery time-based metabolic response patterns were analyzed by Principal component analysis and Partial least squares Discriminant Analysis. Overall, the results of multivariate statistical analysis demonstrated a number of potential biomarkers that were characterized by metabolicomic approach and provided an insight into quantitative chemical accident damage assessment.

WE175

Influence of soil organic amendments on the phenolic contents in rosemary (Rosmarinus officinalis L.) plants

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Rosemary (Rosmarinus officinalis L., Lamiaceae) is an aromatic shrub native from Mediterranean region, grown as a common herb around the world. This species constitutes an exceptional source of different bioactive compounds, mainly phenolic compounds, with proved antimicrobial and antioxidant activity. Furthermore, different studies have shown the potential and important role that this shrub can have in the rehabilitation of degraded soils such as agricultural ones with low levels of organic carbon, contributing to the reduction of erosion and improving soil quality. Within this context, different studies have shown that the metabolism of the phenolic compounds in plants has been associated to environmental factors, such as temperature, rainfall and ultraviolet radiation incidence, as well as soil composition. In this sense, plant nutrient balance in the soil could influence the production of secondary compounds, the concentrations of secondary metabolites in the tissues or the concentration and availability of the necessary nutrients in the soil. Therefore, this study evaluates the effectiveness of the rosemary plant to improve soil quality and the effect of the incorporation of two composts derived from anaerobic digestates on the phenolic contents of rosemary plants grown a semiarid soil. In the study, two composts (CM, mainly composed by cattle manure anaerobic digestate and CS, mainly composed by pig slurry anaerobic digestate at a rate of 30 t ha⁻¹ and 40 t ha⁻¹ respectively) were incorporated into a semiarid soil from central Italy. These organic amendments were compared with the soil without amendment (control treatment, B) and an inorganic treatment (I). Subsequently, plants of rosemary (Rosmarinus officinalis) were planted on these soils. The efficiency of the treatments was evaluated by analysing chemical characteristics in the soil and the total contents of phenolic compounds and flavonoids in the rosemary plants grown in the different treatments. The results obtained have shown that the incorporation of the organic amendments into the semiarid soil improved soil characteristics, by increasing organic matter and nutrient contents, but also implied a decrease in the concentrations of phenolic compounds in the rosemary plants, probably due to the nitrogen fertilisation increases growth, but also leads to decreased concentrations of carbon-based secondary metabolites, such as phenolic compounds.
Freshwater and Marine Ecology

The European Union Water Framework Directive does not require member states to monitor sediment quality. When performed at all, water authorities most often monitor sediment quality by means of chemical target analysis focusing only on target compounds, potentially overlooking ecotoxicological risks caused by (un)known mixtures of sediment associated compounds. Hence, there is an urgent need to incorporate effect-based monitoring and chemical fingerprinting into sediment quality assurance. Therefore, the aim of the present study was to innovate ecotoxicological sediment quality assessment by incorporating whole sediment bioassays and chemical fingerprinting of bioavailable compounds. To this purpose intact whole sediment cores were collected using a sediment core sampler at a reference and 11 contaminated sites grouped by land use: urban, agricultural, and fastwater (WWTP) dominated sites. A Chironomus riparius 28-day life cycle whole sediment bioassay was performed with survival and emergence as endpoints. Simultaneously, SPME fibers were applied as sediment passive samplers to determine pore water concentrations of phenanthrene and pyrene, selected as model compounds for sediment PAH concentrations. Survival in the bioassay was unaffected at the urban sites, while significantly lower at all WWTP sites and two of the agricultural sites. Emergence was significantly delayed at the urban sites, agricultural sites exhibited an irregular emergence time, while WWTP sites induced accelerated emergence. Pyrene and phenanthrene concentrations were negligible at the reference site, very low at the agricultural and WWTP sites, and highest at the urban sites. Urban sites thus have a high chemical load, but survival was higher than on the agricultural sediments. Contrastingly, aggregation was lower at the WWTP and increased at the urban sediment bioassays. From the 49 chemicals included in the dataset, 28 were significantly related to the outcome of Daphnia magna and Chironomus riparius. The Species Sensitivity Distribution (SSDs) method was used to quantify the ecological risk associated with concentrations of contaminants. Based on the SSDs the potentially affected fraction (PAF) of species was calculated. These PAFs were used to calculate the multiple substances PAF, combining effects posed by multiple compounds. Such correlation analysis have not been conducted previously for a large dataset of field-collected sediments in the Netherlands. With our work we contribute to the quantification of relationships between chemical concentrations and toxicological assay in sediments. These relationships can be used in future analyses of predictive abilities of sediment quality and can be applied in an assessment tool for sediment management to determine management strategies.

WE182

Integrative approach to assess ecological risks of sediment metallic contamination in Lake Ohrid (Albania)

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It is now widely admitted that chemical monitoring of pollutants in waters and sediments is not sufficient to assess the risks caused by such pollution in aquatic ecosystems, since chemical data alone provide no indication of biological effects. Biological responses of exposed organisms need to be taken into account, allowing to define the ecotoxicological status of the studied system. The ancient lake Ohrid, the largest lake in the Balkan peninsula, is located between Albania and Macedonia, chosen as case study. The Albanian side, due to the presence of ultramafic rocks, was a large mining area exploited to produce nickel, chromium, and iron until the early nineties. Several ore dumps from this past activity still remain near the shoreline, representing one potential input source of these metals. Several creeks flowing across soils naturally rich in metals also contribute to metal inputs into the lake. We studied three sites along the Albanian shoreline of Lake Ohrid, and defined by different metal pressures: “Pog” in an urbanized area but considered as dimly contaminated by metals, “Menn” and “Poj” located in the ultramafic area of the lake, at the vicinity of a Fe-Ni dump site for “Menn”, or nearby the outlet of a creek for “Poj”. In the two sites under metallic pressure, sediments contained high levels of metals with concentrations reaching 93.8 mg/kg for Co, 345.1 mg/kg for Ni, 335.8 mg/kg for Cd, 133.7 mg/kg for Cu, and 872.9 mg/kg for Fe. Despite these high concentrations, metals are not necessarily bioavailable. This is why it is also important to combine chemical characterization (total and available pools) with the study of lethal and sublethal effects after acute and chronic exposure. In our study, we assessed ecological risks in Lake Ohrid using an integrative approach consisting in: (1) chemical and physical characterizations of sediments, (2) assessment of metal bioavailability, (3) ecotoxicological bioassays, and (4) the study of sub-lethal effects on organisms. During this presentation, the main results from this integrative work at Lake Ohrid will be presented.

WE183

Active Biomonitoring and DGT Passive Sampling: Holistic Assessment of metal bioavailability in sediments and associated risks

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Impacts of sediment metal contamination on aquatic ecosystems and their functioning remain a widespread problem. The ecotoxicological risk associated with metal contamination is dependent on metal speciation, sediment characteristics and the behavior and physiology of the affected organisms. Hence, bioavailable concentrations, rather than total metal concentrations, are often the critical factor in sediment risk assessment. Determination of bioaccumulation in organisms is a frequently used indicator for bioavailability. However, active and passive biomonitoring techniques are often time consuming and highly dependent
on the exposed organisms, limiting comparability and standardization. Diffusive gradient in thin films (DGT) passive sampling is an innovative technique, allowing for the time-integrated measurement of potentially bioavailable metals in sediments or surface water. Divalent metals are selectively accumulated onto a Chelx-embedded hydrogel layer, providing a measurement of labile and weakly-bound metals. To evaluate DGT passive sampling measurements as a potential indicator of bioaccumulation in organisms, a field experiment will be carried out in April 2018 on 6 locations in Flushing Bay, New York, using a freshwater and 3 brackish aquatic systems. Bioavailability of metals will be assessed by active biomonitoring through a 4 weeks exposure of caged macroinvertebrates, after which bioaccumulation will be determined. The organisms will be exposed both at the sediment water interface and in the water column. During a pilot study, carried out in November 2017, 3 bivalves and a polychaete worm are exposed in the Zenne river (north of Brussels) to test for their active biomonitoring applicability. During the 4 weeks exposure period, DGT passive samplers will be deployed 3 times for a period of 24h at the sediment water interface and in the water column. Relationships will be determined between the bioaccumulation in the exposed organisms and the determined DGT fluxes. Bioaccumulation and passive sampling measurements as the sediment water interface and in the water column will be evaluated. The experiment aims at establishing relationships between bioaccumulation in different macroinvertebrate species and passive sampling measurements, and further validating the DGT passive sampling technique as a monitoring tool for sediment quality assessments in both freshwater and brackish aquatic systems.

WE184
Biota
turbation in contaminated sediments: effects on exposure, toxicity and biogeochemistry.
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Sediments are a major sink for a range of contaminants. Organism-sediment interactions such as bioturbation can alter sediment physicochemistry, and facilitate the diffusion of reactive chemical species (e.g. O₃) into deeper sediments, potentially changing the oxidation state of various redox-sensitive materials and the fate and toxicity of contaminants. We applied multidisciplinary to (i) characterise influences of bioturbation on contaminant fate, exposure and toxicity to aid current sediment quality assessment; and (ii) assess the potential use of bioturbation in the management and natural recovery of heavily degraded sediment ecosystems. Increased bioturbation in predominantly metal-contaminated sediments increased bivalve (Tellina deltoidalis) and amphipod (Victoriopsia australiensis) survival from 53% to 100% and 42 to 93%, respectively; and reproduction in a second amphipod (Melita plumulosa) from 3 to 65%. This was attributed to the decreased concentrations of dissolved copper in the overlying water associated with bioturbation. Conversely, increased bioturbation in sediments contaminated by metals and hydrocarbons decreased reproduction (44 to 23%), which was attributed to an increased release of polycyclic aromatic hydrocarbons (PAHs).

High-resolution chemical imaging dissolved oxygen and metals in a legacy contaminated sediment mesocosm with a bioturbator present showed the incorporation of copper and zinc by waters within the bioturbated zone. Significant organism-induced fluxes of nickel and zinc into burrow and overlying waters. The presence of nickel and zinc in burrow and overlying waters demonstrated that organism exposure is likely to be greater from the burrow waters than from the pore waters. This is consistent with increased accumulation of zinc observed with co-habitation of bivalves and amphipods. Low copper and lead concentrations in burrow waters during bioturbation events was consistent with the results of previous tests, where copper concentrations were lower in the presence of high bioturbation intensities, possibly due to binding with iron-(oxy)hydroxide phases or to resuspended particulate phases. These results highlight the importance of considering organism-interactions during sediment quality assessments, and the contributions they have to biogeochemistry and contaminant exposure to surrounding ecosystems.

WE185
The diffusive gradients in thin films (DGT) technique predicts toxicity of nickel contaminated sediments to a marine amphipod

Mining of lateritic nickel ore deposits within the Southeast Asia and Melanesia region is expected to intensify as sulphide nickel ore deposits become depleted. The close proximity of these mining operations to coastal ecosystems places marine benthic organisms at a potential risk of adverse effects related to nickel exposure. Currently, limited data exists for the effects of sediment nickel exposure on coastal marine organisms. The diffusive gradients in thin films (DGT) technique has emerged as a tool that allows for the rapid in situ measurement of the lability and dynamics of metals in sediment. The objective of this research was to determine effects thresholds for sediment-nickel by measuring reproduction of the estuarine-marine amphipod, Melita plumulosa in 10-d whole-sediment bioassays with three nickel-spiked sediments and two field-collected nickel-contaminated sediments with varying chemical and physical properties. We compared the consistent toxic response to both the nickel concentrations obtained using traditional methods of metals from sediments with DGT-labile nickel to determine whether DGT can be used to predict nickel bioavailability and toxicity. Effect concentrations of total recoverable nickel (TR-Ni) to cause a 50% impairment in reproduction (EC50) were 2000 (1200-2900), 1100 (580-1700) and 1100 (740-1500) mg/kg for the silty, sandy-silt, and sandy sediments, respectively. Concentration-response relationships based on DGT-labile Ni fluxes had less variation and better-predicted toxicity in the field collected nickel-contaminated sediments. Site 1 (2000 mg/kg TR-Ni) and Site 2 (1300 mg/kg TR-Ni) had reproductive responses of 88% ±10 and 71% ±11 of the control, respectively. The EC50s based on DGT-labile Ni were 2.3 (1.7-3.4), 3.3 (1.7-4.9) and 2.0 (1.0-3.0) mg/m³ for silty, sandy-silt and sandy sediments, respectively. Concentration-response relationships based on DGT-labile Ni fluxes had less variation and better-predicted toxicity in the field collected nickel-contaminated sediments. Site 1 (0.4 mg/m³ DGT-labile Ni) and Site 2 (1.0 mg/m³ DGT-labile Ni) sediments, respective reproductive responses were 88% ±10 and 71% ±11 of the control. This demonstrates that amphipods were responding to the labile nickel as measured by DGT and further supports its use in nickel risk assessments.

WE186
Identifying key toxicants in sediment samples from urban waterways in Guanzhou, China using a integrated method of TIE and EDA
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Use of a triad approach to assess their influence on sediment quality assessment. Two approaches, namely toxicity identification evaluation (TIE) and effect-directed analysis (EDA) have been developed. Conventional sediment TIEs take the advantage of environmental relevance by using whole organism bioassays while suffer from lack of effective methods for specifically identifying major contaminants from a universe of chemicals of organic contaminants are identified as main class of toxicants in phase I TIE. Alternatively, EDA is a powerful tool in identifying causes of sediment toxicity with sophisticated fractionation and chemical analysis targeted of non-targeted toxicants, but it is short of environmental relevance due to the use of in-vitro bioassays and exhaustive solvent extraction. To better understand the cause of sediment toxicity in urban waterways in Guangzhou, China, a new methodology combining the potential use of bioturbation in the management and natural recovery of heavily degraded sediment ecosystems. Increased bioturbation in predominantly metal-contaminated sediments increased bivalve (Tellina deltoidalis) and amphipod (Victoriopsia australiensis) survival from 53% to 100% and 42 to 93%, respectively; and reproduction in a second amphipod (Melita plumulosa) from 3 to 65%. This was attributed to the decreased concentrations of dissolved copper in the overlying water associated with bioturbation. Conversely, increased bioturbation in sediments contaminated by metals and hydrocarbons decreased reproduction (44 to 23%), which was attributed to an increased release of polycyclic aromatic hydrocarbons (PAHs).

High-resolution chemical imaging dissolved oxygen and metals in a legacy contaminated sediment mesocosm with a bioturbator present showed the incorporation of copper and zinc by waters within the bioturbated zone. Significant organism-induced fluxes of nickel and zinc into burrow and overlying waters. The presence of nickel and zinc in burrow and overlying waters demonstrated that organism exposure is likely to be greater from the burrow waters than from the pore waters. This is consistent with increased accumulation of zinc observed with co-habitation of bivalves and amphipods. Low copper and lead concentrations in burrow waters during bioturbation events was consistent with the results of previous tests, where copper concentrations were lower in the presence of high bioturbation intensities, possibly due to binding with iron-(oxy)hydroxide phases or to resuspended particulate phases. These results highlight the importance of considering organism-interactions during sediment quality assessments, and the contributions they have to biogeochemistry and contaminant exposure to surrounding ecosystems.

WE187
Water discharges from the city of Lausanne during rainfall in Lake Geneva: Use of a triad approach to assess their influence on sediment toxicity
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To do this, a sampling grid composed of 15 sites was developed in the discharge area of the effluent from the Flon river into the lake. At each point, sediment samples were collected to measure metal concentrations and assess the ecotoxicological quality of sediments in the laboratory using a whole sediment toxicity test with ostracods. At six selected sites in the central transect of this sampling grid, corresponding to the extension of the outlet of the Flon river, a more detailed monitoring program was applied, with measurements of PCBs and PAHs concentrations. During the experiments, the chemical toxicity tests with chironomids, macrophytes and nematodes, as well as the study of the structure of oligochaete communities and tolerance of microbial communities induced by pollution (PICT) were carried out on. The results obtained showed that contamination induced by urban stormwater discharges, identified mainly as copper, zinc, PCBs and PAHs contamination, was elevated in the sediments at certain locations. The results of these experiments also did not indicate significant toxicity in this area, the study of in situ communities revealed the presence of pollution-resistant species among oligochaetes and benthic microorganisms. For the most remote sites, the effects observed in the sediments do not appear to be directly related to the stormwater discharges. The origin and dynamics of the contamination still require further investigations, based in particular on a hydrodynamic model. Overall, this study paves the way for the development of practical tools for assessing the impacts of urban stormwater discharges in lakes in Switzerland.

WE188
Ecotoxicological profiling of sediments along the River Wurm by Aachen (North-Rhine-Westphalia, Germany)
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River sediments serve as a sink and source for micropollutants. Characterized by their semi-dynamic behaviour, sediments can assimilate contaminants. Naturally occurring events such as storms, currents and flood events, as well as human activities like dredging can cause resuspension of sediments and, thus, pose a threat to aquatic organisms. So far, many investigations have been conducted to assess the biological responses in the water phase of streams being impacted by effluent from waste water treatment plants (WWTPs). However, the impact of WWTPs to sediments is still unknown. The present study was taking place within the DemoAC-AC project aimed to assess the ecotoxicological status of the Wurm River near Aachen. This stream is heavily impacted by anthropogenic activities, such as urbanisation, agriculture, industry, etc. Moreover, the River Wurm receives effluents from various WWTPs within the catchment area. Two of them, the WWTP Aachen Soers and the WWTP Ellendorf, served as investigative objects. Special attention is paid to the WWTP Aachen Soers, which will be upgraded by a full-scale ozonation at the end of 2017. To evaluate a possible impact of the WWTPs on the current ecotoxicological status of the stream a comprehensive ecotoxicological profiling of the sediments from 7 sites up- and downstream of the aforementioned WWTPs was performed. The samples were tested both as native and freeze-dried samples in the sediment contact assay with Danio rerio. Sediment extracts (25 ± SEQ/ml) were applied for the fish embryo toxicity test with Danio rerio as well as for the determination of the Caco-2 cell uptake and the mutagenic potential. The results showed estrogenic and mutagenic potential in sediment extracts upstream the tested WWTPs. The embryo toxic potential (enlarged heart, insufficient blood circulation, oedema, etc.) was accounted for the sediment extracts upstream of the WWTP Aachen Soers only. Exposure to native sediments did not lead to any adverse effects in embryos of the zebrafish. However, exposure to freeze dried sediments revealed reduced reactivity of fish embryos. Observed neurological conspicuousness will be verified by further investigations. The described toxicological profiling of sediments will also be completed by chemical analysis. Phase 2 of the DemoAC-AC-project will contain comparative studies in order to evaluate the possible influence to sediment toxicity after implementation of full-scale ozonation.

WE189
Comparing conventional and integrative concepts for sediment classification systems
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Environmental regulations and guidelines in Europe for assessing the quality of aquatic sediments and dredged material predominantly demand chemical data, and decision making mostly does not integrate the information from different lines of evidence (1, 2). Ecotoxicological data requirements are often limited, with the final classification of the sample not preserving the information of all applied biotests (3). Improved, holistic characterization of sediments and dredged material is needed, to enable a better risk assessment that conserves the ecological quality, is practical and economically feasible at the same time. This poster will present the concept of a study in the scope of the Interreg project “Sullied Sediments” (http://northsearegion.eu/sullied-sediments/) and will discuss first results. The study aims at comparing and evaluating conventional and alternative, integrative and science-based sediment classification concepts for holistic assessments of sediment quality, such as fuzzy-logic based classification (4, 5). Selected concepts will be applied on the classification of sediments from inland waterways in the North Sea region. A sediment quality trial approach will assess the ecotoxicity, the ecophysiological status and the chemical composition of the sediment samples from this catchment area of the North Sea region with management problems related to polluted sediments. The locations of the sites are in the Port of Hamburg and along the River Elbe in Germany, on the Rivers Hull in the UK and Scheldt in Belgium and in the Netherlands. The applied biotest battery for assessing the ecotoxicological potential of the sediments will cover different trophic levels, different sensitivities and different exposure pathways, considering the toxicity of sediment contact tests, eluates and extracts. The analysis of the benthic meiofaunal community will assess the ecology of the sediments. The chemical analyses will comprise a broad range of historic contaminants and emerging pollutants, originating from industrial activities, agriculture and pharmaceuticals. This work will be the basis for developing an improved, integrated sediment classification system. References: Ailh et al., 2002. JSS 2: 37–42 Deckere et al., 2011. JSS 11: 504–517 Duff et al., 2003. Federal Ministry for the Environment, Nature Conservation and Nuclear Safety (BMU) Hollett et al., 2002. Ecotoxicology 11: 311-321 Keiter et al., 2009. JSS 9: 168

WE190
Submarine sewage outfall adversely affects the sediment quality of Santos, Brazil estuary - An acute toxicity study
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The submarine sewage outfall of Santos, Brazil is an important facility for sewage disposal in several countries is primary treatment followed by chlorination and its discharge in the sea. In Santos city, SP, Brazil, 1 million of cubic meters of urban effluent are discharged into the Santos Bay every day, 4,5 km from the beach. To assess the toxicity of environmental samples it is important to use species that are representative of the ecosystem we want to protect. So, the aim of this work was to evaluate the acute toxicity of water and sediment samples collected in the area under the influence of this discharge using the native marine amphipod Parhyale hawaiensis. Three campaigns were conducted. Acute toxicity tests were performed in water and in fresh and dried sediment as well water and sediment extracts. The fish used was Danio rerio and Parhyale hawaiensis. The results obtained showed that contamination induced by urban stormwater discharge was toxic ranging from 17 to 100% mortality as well the respective controls. Fresh and dried sediment were toxic ranging from 17 to 100% mortality as well the respective organic extracts. The observed toxicity is probably mainly related to organic contaminants adsorbed to the sediment particles. The sediment of the area seems to be adversely affected by the influence the outfall discharge. Acknowledgements: FAPESP 2015/24758-5 and CNPq 400362/2014-7

WE191
Swimming in turbid water: impacts of suspended fine sediments on fish physiology
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The impact of sediments on aquatic ecosystems is significant, mainly due to their high organic content. Anthropogenic activities lead to increasing sediment deposition in many rivers worldwide which must be managed to preserve industrial activities and population safety. In this context, swimming and daphnia diurnal activities are common actions that release downstream accumulated sediments thus increasing sediment loads which in turn impairs freshwater biodiversity. Reported impacts on fish species vary from mortality, behavioral changes, to physiological and histological impairment depending on SSC and exposure duration. Juveniles of rainbow trout (Onchorhynchus mykiss) and roach (Rutilus rutilus) were exposed in semi-static conditions with 0, 5, 10, 20 and 1000 mg/L of non-contaminated fine sediments (microna) for 28 days, mimicking dredging operations in terms of duration and environmental concentrations. They were sampled weekly, and mortality, condition index and neurophysiological parameters were also investigated to assess the level of oxidative stress and genotoxicity. Oxidative damages in gills were investigated measuring the level of lipid peroxidation (TBARS) as well superoxide dismutase activity (SOD) involved in oxygen radical metabolism. The level of primary DNA damage in erythrocytes was measured with the alkaline comet assay. This preliminary work highlight that 28 days of exposure to fine sediments at high concentrations do not induce drastic
mortalities as predicted by models and the scientific literature. Physiological parameters investigated in rainbow trout acted as early signals of biological defects pointing out a high level of genotoxicity measured in erythrocytes in exposed individuals as well as in the control batch; these decreased during the experiment until a basal level pointing out the resilience of fish whereas they were exposed for 28 days to high fine sediment concentrations. Roach exposure to suspended fine sediment did not cause genotoxicity or an oxidative stress. These results meant that fine sediment exposure did not lead to a physiological stress through the alteration of respiration and osmoregulation homeostasis but suggested that trout experienced undesired past stressful conditions (aquaculture) independent from the sediment exposure. However, we cannot conclude that exposures of juvenile fish to such sediment concentrations would not lead to biological detrimental effects without further considering environmental sediment quality.

**WE192 Assessing the bioavailability of metals in natural sediments by DGT passive sampling and bioaccumulation**

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Worldwide, high metal concentrations from recent and historic sediment contamination form a widespread problem and are of major concern for water system management due to their impact on the surrounding water quality and resident biota. Sediment (metal) microorganisms can be present in a broad range of physiochemical forms, some of which may be unavailable, non-toxic and therefore not-harmful to organisms so that the interplay between chemical speciation and biological effects can be very site specific and hard to predict. Total sediment concentrations are therefore often found to be poor predictors of the actual risk and a measure of bioavailability should be considered in risk assessment procedures. In this regard low-level low-level techniques such as the DGT bioaccumulation in exposed organisms has been experienced to be highly disruptive, time consuming and limited in comparability as its results are strongly dependent on the analysed organisms themselves. Therefore, an increasing need for less disruptive, more reliable and standardized methods exists. Recently, passive samplers have been tested to estimate bioavailable contaminant fractions as well as the contaminant flux over sediment-water interface. Diffusive Gradient in Thin film samplers (DGTs) have been indicated to provide reliable predictions of metal bioavailability and toxic potential for single (benthic) invertebrate species under (semi-) controlled conditions. The main objective of this study is to further evaluate the use of DGT passive samplers as indicators for the bioavailability of metals for (benthic) microorganisms and to test the robustness of the results from laboratory studies under field conditions. In an extensive field and laboratory study, which will be performed in April 2018, the impact of a range of contaminated natural freshwater sediments with known physicochemical characteristics and metal gradients on species performance will be tested. Bioavailable metal fractions will be determined by the use of DGTs and by measuring the metal body burden and metal concentrations in microorganisms. We assume that results from these experiments are expected to increase the insights in the applicability of passive samplers for future sediment risk assessment and to be useful for the development of more standardized and integrated approaches.

**WE193 Ecotoxicological effects of sediments influenced by a municipal wastewater treatment plant - state of a receiving river before implementing an ozonation treatment**

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Since our economic progress continues, the environmental pollution increases. The contamination of the aquatic environment with chemicals is one of the major consequences of our consumption society. Monitoring 500 million tons of compounds are applied in industrial processes and consumables every year. A considerable part of it enters waterbodies from diverse and point sources. [1] Micropolutants originating from e.g. pharmaceuticals and personal care products may cause adverse effects on different biological and ecological levels. A major concern is the fact that these substances are not fully removed during common wastewater treatments and, therefore, are found in surface discharge of micropolutants from wastewater treatment plants (WWTP) additional treatment steps are required. Ozonation has been shown to be an effective method with reasonable costs. Hence, ozone treatment of the entire effluent is implemented in the Aachen-Soers WWTP, Germany, within the DemoAC-project. In this context, the actual ecotoxicological state of the recipient water, the River Wurm, and the upstream tributary, the River Haarbach, was evaluated before the implementation of the facility. The River Haarbach receives effluents from the Aachen-Elidennicht WWTP. Therefore, water and sediment samples from various sites upstream and downstream the WWTPs were investigated. This study focuses on the assessment of the sediment samples of the both rivers, since they play an important role in e.g. binding and remobilisation of substances. After the extraction of the sediments via a pressurised liquid extraction, cell-based bioassays with reporter cell lines will be conducted to estimate the anti-estrogenic and oxidative stress potential. Following, both native samples and extracts will be tested in the behavioural light/dark transition test with Danio rerio. This test utilizes the fish’s scototaxis (aversion to bright areas and natural preference for the dark) to evaluated effects of neuroactive compounds within these matrices. [1] Schwarzenbach et al. (2006).

**WE194 Dredging sediment quality evaluation: a comparison of an ecotoxicological classification using an weight-of-evidence approach and a “pass to fail” criteria**

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Recently a new regulation for the management of dredging sediment has been introduced in Italian legislation (Decree of Italian Ministry of Environment n. 173/2016), establishing criteria and methodological procedures for dredging sediment characterization, their classification and identification of appropriate management options and monitoring. One of main novelties is represented by the Vicinuity scale, allowing the evaluation of a specific sediment sample based on the results from laboratory-based bioassays. The scale is composed by four main steps: the test with aquatic microalgae Chlamydomonas reinhardtii, the test with the crustacean Daphnia pulex, the test with the insect Chironomus dilutus, and the test with the fish species Carassius auratus. A battery of bioassays that considers the use of three species belonging to different trophic levels of aquatic ecosystems and the organisms has been shown to be highly robust, time consuming and limited in comparability as its results are strongly dependent on the analyzed organisms themselves. Therefore, an increasing need for less disruptive, more reliable and standardized methods exists. Recently, passive samplers have been tested to estimate bioavailable contaminant fractions as well as the contaminant flux over sediment-water interface. Diffusive Gradient in Thin film samplers (DGTs) have been indicated to provide reliable predictions of metal bioavailability and toxic potential for single (benthic) invertebrate species under (semi-) controlled conditions. The main objective of this study is to further evaluate the use of DGT passive samplers as indicators for the bioavailability of metals for (benthic) microorganisms and to test the robustness of the results from laboratory studies under field conditions. In an extensive field and laboratory study, which will be performed in April 2018, the impact of a range of contaminated natural freshwater sediments with known physicochemical characteristics and metal gradients on species performance will be tested. Bioavailable metal fractions will be determined by the use of DGTs and by measuring the metal body burden and metal concentrations in microorganisms. We assume that results from these experiments are expected to increase the insights in the applicability of passive samplers for future sediment risk assessment and to be useful for the development of more standardized and integrated approaches.

**WE195 Toxicity of sediment-bound lufenuron to aquatic arthropods in laboratory bioassays**

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Lipophilic pesticides are frequently detected in sediments, potentially leading to toxic effects on benthic organisms. Currently, prospective sediment risk assessments for pesticides are mainly based on results of laboratory bioassays with a few standard test species (Chironomus sp. and Hyalella azteca). It is, however, uncertain whether these standard benthic test species are representative for a wider array of freshwater benthic organisms. We selected the benzoylurea insecticide lufenuron as one of the benchmark substances to evaluate the prospective environmental effect assessment procedure for sediment-associated pesticides. 10-day and 28-day toxicity estimates from sediment-sampled laboratory bioassays with benthic arthropods belonging to different taxonomic groups are presented. In the 10-d sediment-sampled toxicity tests the LC50 values showed the following order from low to high LC50: Caenis horaiia > Chironomus riparius > Gammarus pulex > Chironomus dilutus > Daphnia pulex > Hyallela azteca > Asellus aquaticus and Sialis lutaria. The Hazardous Concentration to 5% of the tested species (HC5 and 95% confidence limit) derived from these 10d-LC50 values was 2.2 (1.2-5.7) µg/g organic carbon (OC). This HC5 value is approximately a factor of 2 lower than the 10-d LC50 estimate (4.37/µg/g OC) for the most sensitive standard test species Chironomus riparius. Valid 28-d LC10 values could be derived for 7 benthic arthropods. These 28d-LC10 values showed the following order from low to high LC10: Asellus aquaticus > Chironomus riparius > Caenis horaiia > Ephemera danica > Hyallela azteca > Gammarus pulex > Sialis lutaria. The HC5 and 95 confidence interval derived from these 28d-LC10 values was 0.13 (0.02-1.30) µg/g OC. This HC5 value is approximately a factor of 3 lower than the 28-d LC10 estimate for the most sensitive standard test species Chironomus riparius (0.49/µg/g OC). These data show that Chironomus riparius is a representative standard test species to assess the potential risks of sediment exposure to the insecticide lufenuron. The HC5 obtained from 28-LC10 values was a factor of 6 lower than the the NOEC for the most sensitive
population (0.79 µg/g OC) in a sediment-spiked microcosm experiment, while the HC5 from 10-d-LS50’s was approximately a factor of 3 higher than this microcosm threshold concentration.

WE196
Application of an undisturbed sampling technique for depth related analysis of aquatic animals to OECD TG 219 sediment test systems
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Sediment toxicity testing of plant protection products (PPP) is gaining an increasing awareness within the scientific and regulatory community. Currently, PPP concentrations in sediment and pore-water of Chironomid toxicity tests acc. to OECD test guideline (TG) 218/219 are determined as mean over the entire sediment layer of the test system. Hence, a depth-related measurement would contribute to a more accurate assessment of the effective exposure for the predominantly surface sediment dwelling test organism. Therefore, we developed an undisturbed sampling technique and processing enabling the depth-related analysis of active substances in pore-water and adsorbed to sediment particles. After removing the water phase, plastic tubes were stung into the ca. 15 mm-thick sediment layer, which was subsequently frozen by liquid nitrogen. By the use of a special developed cutting device, the sediment cores were cut into three slices providing a thickness of ca. 5 mm. Each sediment slice was centrifuged to isolate the pore-water. A sequential extraction was performed to extract the sediment adsorbed residues. After combining the sediment extracts, pore-water and sediment extracts were analysed by LC-MS/MS. To validate this newly developed sampling technique, a Chironomus toxicity study acc. to OECD TG 219 was conducted. Two model compounds (A, logP = 1) and B (logP = 3) were applied as mixture at nominal concentrations of 2 µg/L. To investigate the spatiotemporal behaviour of the compounds, test systems were incubated and processed 3, 7, 14, 21, and 28 days after treatment (DAT). The concentration of the applied compounds decreased in the overlying water during the experimental duration. Both compounds were primarily found adsorbed on the sediment phase (ca. 40-50 % of applied compounds). Approx. 0.2 - 1 % of the applied compounds were recovered in the pore-water at the same time. The analytical results of pore-water and sediment extracts show that the highest amounts of both compounds were in the upper layer of the sediment during the experimental duration. Nevertheless, the results indicate the concentrations differences between the top layer and the layers below will be equalized in the course of time. These first results indicate that the newly developed sampling technique can provide a substantial contribution to a more realistic determination of exposure concentrations in chronic water-sediment toxicity tests, leading to an improved sediment risk assessment.

WE197
SETAC Sediment Interest Group
P.K. Sibley, University of Guelph / School of Environmental Sciences
Improving the environmental risk assessment of the aquaculture 'Blue Revolution' (P)
WE198
Global overview of aquaculture production with a focus on the development and current status of the activity in Portugal
C.V. Rocha, MARE-FCUL; H.N. Cabral, Faculdade de Ciências da Universidade de Lisboa / MARE-FCUL; J.C. Marques, University of Coimbra / MARE, Dep. of Life Sciences, Coimbra University/Biologia Department & CESAM, Aveiro University/Aquaculture activity experienced true global development firstly at the beginning of the 1990’s and then around 1970’s, as a result of the awareness of the negative impacts that years of intensive fisheries brought to wild stocks’ status, which contributed to overexploitation. 

Antibiotics in Aquaculture
Antibiotics have been used in aquaculture since the 1940s for treatment of diseases, improving the survival rates of fish and crustaceans. Their use has increased significantly in recent decades, particularly in intensive rearing systems. While antibiotics can help mitigate disease outbreaks, their widespread use in aquaculture has raised concerns about antibiotic resistance, environmental impact, and public health implications. 

Aquaculture practices, especially concerning the control of water quality and animal feeding, in the past decades, however, extensive and semi-intensive rearing systems are poorly characterized, raising questions not only about the influence of environmental factors on production, but also on the threats that the activity may pose to the surrounding environment. Among these are, for example, the destruction of natural habitats due to facilities extension and aquaculture effluent discharges with high nutrient input to the surrounding water bodies. Careful site selection and efficient waste management plans are imperative to minimize these potential threats of aquaculture practices. Although fish supply for human consumption from aquaculture has already surpassed that of fisheries, concerns about farmed fish quality have been raised. Fortunately, it has also resulted on the honing of aquaculture methods and practices, especially concerning the control of water quality and animal feeding, in order to achieve the highest quality product.

WE199
Characterization of the ontogenetic variation and nutritional composition of Gilthead seabream and European seabass reared in two Portuguese estuaries
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The nutritional value and potential ontogenetic variations of cultured fish was assessed for the European seabass and the gilthead seabream species reared in semi-intensive systems in two southern European estuarine systems, the Sado and Mondego estuaries in Portugal. Quantification of total protein, carbohydrate and fatty acid profiles were determined to assess differences between the organoleptic composition of organisms of the same species reared in four different aquacultures (two estuaries in each country). No significant differences were found among groups of both species regarding protein content. A significant influence of the rearing site was found for the European seabass regarding saturated (SFA), monounsaturated (MUFA) and highly unsaturated fatty acid (HFA) contents, either between estuaries and within each estuary. In the Gilthead seabream, SFA, MUFA, polyunsaturated fatty acid and HFA contents were also influenced by the rearing site. Eicosapentaenoic acid, docosahexaenoic acid, arachidonic acid and linoleic acid content in adults specimens were dependent on the fish rearing site. In general, seabass and seabream fatty acid content was higher in organisms reared in the Sado estuary, when compared to the individuals reared in the Mondego estuary. Carbohydrate analysis showed a significant influence of the rearing site on free sugars and polysaccharide content in fish of both species. Differences in fatty acids and carbohydrates content among juvenile and adult stages were found for all groups studied. The present work supported the existing evidence that semi-intensive rearing systems are subjected to the variability of extrinsic factors, namely the different anthropogenic pressures these systems are subjected to, causing fluctuations in water quality and composition, which may influence the fish growth. From a consumer’s perspective, such differences may come as a disadvantage, as it is expected for a product to provide equal nutritional properties and benefits regardless its origin, especially within the same country. From the farmer’s perspective, such vulnerability requires constant monitoring of the extrinsic parameters within the facilities, in order to avoid great losses of the product. Further studies should be conducted to define the parameters that should be regulated and monitored in semi-intensive systems to obtain the best product with the highest food quality.

WE200
Effects of aquaculture antibiotics on marine biofilms and on the amphipod Gammarus aequicostatus
Immune aquaculture is considered to be an important source of antibiotics into the marine environment. Antibiotics may cause adverse effects on potentially sensitive organisms such as non-target microorganisms, primary producers or benthic invertebrates. The objective of this study was to evaluate the potential side effects of two antibiotics used in aquaculture (oxytetracycline and fluencequim) on the community composition of marine biofilms exposed to these substances and on the survival of Gammarus aequicostatus. Marine biofilms were exposed to 1, 10, 100 and 1000 µg/L of oxytetracycline and fluencequim for one week under laboratory conditions. Subsequently, the exposed biofilms were used to feed G. aequicostatus organisms for two weeks. The G. aequicostatus aquaria set up was run with two treatments in parallel: (1) with input of antibiotics only from the biofilm and (2) with antibiotics administered via biofilm and also spiked into the water. This was done to test different bioaccumulation routes (i.e., ingestion only and water exposure combined with ingestion). All the treatments for biofilm and crustaceans test were run in triplicate. Preliminary results show a marginally enhanced biomass growth of the biofilm with increasing dose of both antibiotics,
being this slightly higher in the oxytetracycline test. No correlation was found between antibiotics concentration and elemental composition (analyzed carbon, sulfur and total phosphorous), although nitrogen content was slightly higher in the medicated biofilm. Biofilm arborescence (vertical structures observed through optical microscopy) coverage was statistically different among treatments, showing a non-linear response. Experimental results show that low exposure concentrations contributed to higher bioerosion up to 20% g/L, while the highest tested concentration contributed to the 60% of the biofilm's decrease. Ongoing work includes the evaluation of antibiotic's bioaccumulation in the biofilms, bacterial genetic characterization (microbiome and resistome), diatoms identification, and photosynthetic activity assessment. Regarding the G. aquicuadua test, the experimental results do not show an effect over their size or weight by the feeding on antibiotic exposed biofilms. Reproduction and survival were not compromised either. Also bioaccumulation will be analyzed to determine the relevance of each of the evaluated antibiotic exposure routes.

**WE201**

**Shifts in the diatom assemblage structure and biological traits of marine biofilms exposed to antibiotics used in aquaculture**

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The use of antibiotics, anti-fouling paints, and fish food are all potential sources of chemical contaminants from commercial aquaculture operations such as fish farms. In particular, antibiotics are added to fish feed to prevent infections or cure diseases, and other compounds such as copper and zinc are added in anti-fouling paint. This exposes the marine environment to these substances. Biofilms were grown during two weeks under natural conditions on glass slides. Two experiment were conducted: one was another laboratory setup and the other experiment field, grown marine biofilms were exposed to 1, 10, 100 and 1000 µg/L of each single antibiotic compound for one week and then changes on chemical and biological composition were analyzed. In the second experiment, biofilms exposed to the same antibiotic concentration range were transported to field conditions after two weeks of exposure in order to evaluate their recovery capacity. In both experiments, diatoms were determined to the lowest possible taxonomic level under the microscope (Nikon Eclipse TE2000-u). The taxonomic abundance of the sampled quadrats of each replicate was averaged and referred to the area sampled to obtain the taxon density per replicate. The fine structure of diatoms was analyzed under a scanning electron microscope (JEOL-JSM-6100). The diatom composition, the relative abundance of species (%), the Shannon-Wiener diversity index (H') and species richness were calculated for each sample and then summarized per treatment. The growth forms (biological traits) of species were analyzed before detachment and were grouped according to the literature. The biofilms were dominated by a reduced number of taxa, including the diatoms Brachysira aponica and Coccones plicatula. High exposure concentrations of oxytetracycline and fluoxetine (100 and 1000 µg/L) resulted in an abundance decrease of the genera Hyalosynedra and Licmophora. The global architecture and traits of the biofilms were also influenced by the high antibiotic exposure concentrations.

**WE202**

**Assessing the oxidizing effects of hydrogen peroxide using flow cytometry as a high throughput method**

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Hydrogen peroxide (H₂O₂) is widely used in commercial, industrial, medical, environmental and hygiene applications. It is applied in aquaculture for controlling biological problems such as salmon lice. H₂O₂ produces highly oxidizing radicals that can cause paralysis, peroxidation in organelle membranes and inhibition of enzymes that replicate DNA in biological organisms. The release of H₂O₂ as an effluent into the marine environment is therefore a cause for concern particularly to primary producers such as algae. With the use of flow cytometry, single cells of algae with different features and physiological states, can be examined based on the fluorescence of species specific probes. The use of probes that demonstrate oxidizing properties enables fluorometric determinations of several parameters such as the ability of reactive oxygen species (ROS) to oxidize non-fluorescent probes to fluorescent products. In the present study, the effects of H₂O₂ on Skeletonema pseudocostatum were analysed. The method provided a rapid assessment of several endpoints in the same exposed samples. Effects on growth, photopigmentations and the detection of intracellular ROS production, using 3 molecular probes, were measured over 72 hours. H₂O₂ was used for determining the oxidative burst, DHR 123 for mitochondrial oxidation, and BODIPYfluorescein to determine lipid peroxidation (LPO). Exposure concentrations were selected to cover the overall concentration response curve and a short-term exposure was also made to discern initial high reactivity of H₂O₂. Chemical analyses were performed to verify the stability of the concentrations during the exposure duration. The short-term exposures demonstrated rapid high toxicity of H₂O₂ to algae, where ROS production and the response to the photopigments were the observed endpoints. Over the 72 h, the response of the algae at the different test concentration clearly differed. The accessory photopigments actively responded when the main natural pigments declined. The ROS protective system seemed to be active at medium concentrations, whereas at higher concentrations damage on membranes lipids and mitochondria possibly instigated cell failure. This high throughput approach demonstrated a great potential to study the oxidizing effects of hazardous compounds in algae. While growth inhibition allowed to discriminate the overall toxicity, the high throughput methods, using flow cytometry, helped to screen and characterize the Mode of Action of H₂O₂.

**WE203**

**An updated version of the SEPA BathAuto tool for assessing anti-parasitic chemical treatments in marine fish farms**

J. Carnall, Cambridge Environmental Assessments; A. Berkeley, Scottish Environment Protection Agency; F. Ericher, CEA; G. Hughes, Cambridge Environmental Assessments

Marine fish farms operators in Scotland wishing to use anti-parasitic chemicals as bath treatments must first obtain a discharge licence from the Scottish Environment Protection Agency (SEPA). Discharge licences are granted by SEPA on a per-site basis, with the quantity of chemical that can be released from a particular fish farm determined by computer modelling. This computer modelling considers the location and composition of the fish farm, along with hydrographic data measured at the site and the toxicity and environmental fate of the chemicals concerned. Discharge quantities are typically calculated for three anti-parasitic chemicals: azamethiphen, cypermethrin and deltamethrin. Of these, cypermethrin and deltamethrin are rapidly removed from the aqueous phase via binding to particles, and are therefore assessed using SEPA’s short-term model, which calculates projected concentrations in the chemical patch up to 6 hours after its release from the fish farm. Azamethiphen, however, remains in the aqueous phase for several days until it is broken down, and is therefore also assessed using a longer-term model, originally developed by Gillibrand and Turrell (1999; MLA Report No 290/99) and recently extended by Carnall, Ericher and Hughes (2017; poster presentation at SETAC Europe 2017). The SEPA tool BathAuto integrates both the short and long-term models, iteratively performing calculations of chemical concentrations in the water in order to arrive at safe discharge limits for a fish farm. In this poster we present an updated version of BathAuto in which the longer-term model is now fully integrated into the BathAuto tool. The standalone executable required previously for the long-term model (openedisp.exe) is no longer used, improving the compatibility of the revised tool with modern operating systems. While the standard modelling parameters required by SEPA for discharge licensing in Scotland are included as default, BathAuto could readily be modified to include alternative parameters or chemical data, potentially making this tool available for use by a wider range of industries. The revised version of BathAuto also offers improved graphical outputs, and the ability to calculate several options for compliant cage treatment regimes.

**WE204**

**State-of-the-art on the use of models for the ERA of chemicals used in aquaculture**


Aquaculture is expanding and becoming a more diverse industry in terms of species and production systems, there is an increasing demand to generate improved modelling tools to assess its environmental impacts. Although important developments have been made in disease prevention and treatment, the use of veterinary medicines and other potentially toxic substances (e.g. antifoulants, metals) is still widespread and used on a large scale. This study presents an overview on the use veterinary medicines and other potentially toxic substances used in EU aquaculture, and the environmental standards and regulatory procedures available for their Ecological Risk Assessment (ERA). Furthermore, it describes the state-of-the-art on the development of models capable of assessing the fate, dispersal, exposure, ecological effects and associated regulatory procedures for their Environmental Assessments. This study shows that a varied range of models has been developed during the last 30 years. Their effective implementation in regulatory ERA is, however, somewhat limited in many state members. Some recommendations are provided as to improve the chemical exposure assessments and the ecological realism of the modelling outcomes, paying a special attention to the protection goals set for the regulatory ERA of veterinary medicines.
populations: from lab experiments to population-level endpoints

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The continued growth of marine aquaculture production has presented the industry with environmental and production concerns, of which the ectoparasitic salmon lice (Lepeophtheirus salmonis) has gradually become a major problem. A commonly used parasiticide against this crustacean is diflubenzuron (DFB), which acts as a chitin synthesis inhibitor and thereby interfere with the molting stages during sea lice development. However, DFB from the fish feed may also affect non-target crustaceans such as the Northern shrimp (Pandalus borealis), which is an economically and ecologically important species in Norwegian fjords. Laboratory experiments have demonstrated that shrimp exposed to DFB through fish feed have reduced survival (ca. 60%) compared to control, in both the larvae and the adult stages. Moreover, the effects of DFB exposure is more severe under future climate conditions (higher temperature). The aim of this study is to make the information on these mechanistic effects more relevant for risk assessment at the population level. We have developed an age-structured population model representing a Northern shrimp population located in a hypothetical Norwegian fjord containing a fish farm, under both ambient and future climates. Our model is based on thorough knowledge of shrimp biology and clear results on toxicological effects from the laboratory experiments. Nevertheless, extrapolating the individual-level effects to the population level poses several challenges. Relevant information on shrimp populations in fjords is sparse (such as abundances, survival and reproductive rates, and their risk-dependent processes). The degree of exposure to potential of DFB at different distances from aquaculture farms is also uncertain. We have therefore developed a set model scenarios representing different medicine application schemes and different degrees of exposure for the shrimp populations. The purpose of the model is to predict effects of DFB exposure on population-level endpoints such as long-term abundance and age structure, and to assess the risk of population decline below threshold abundances.

WE206 Contamination and bioaccumulation of heavy metals in the wild and marine farmed milkfish (Chanos chanos) and mullet (Mugil cephalus) and associated health risk along the coasts of Tanzania
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Concentration of heavy metals Cu, Pb, Fe, Zn, Co, Cr, Cd, Ni and Al were analysed in the muscles and livers of farmed and wild milkfish and mullets from Tanzanian coast. Fish samples were collected from January 2016 to April 2016 and analysed for heavy metals by using Atomic Absorption Spectrophotometer while lead was analysed by using Flame Atomic Absorption Spectrophotometer. The degree of exposure to potential of Fe(lld) at different distances from aquaculture farms is also uncertain. We have therefore developed a set model scenarios representing different medicine application schemes and different degrees of exposure for the shrimp populations. The purpose of the model is to predict effects of DFB exposure on population-level endpoints such as long-term abundance and age structure, and to assess the risk of population decline below threshold abundances. Concentration of heavy metals Cu, Pb, Fe, Zn, Co, Cr, Cd, Ni and Al were analysed in the muscles and livers of farmed and wild milkfish and mullets from Tanzanian coast. Fish samples were collected from January 2016 to April 2016 and analysed for heavy metals by using Atomic Absorption Spectrophotometer while lead was analysed by using Flame Atomic Absorption Spectrophotometer. The degree of exposure to potential of Fe(lld) at different distances from aquaculture farms is also uncertain. We have therefore developed a set model scenarios representing different medicine application schemes and different degrees of exposure for the shrimp populations. The purpose of the model is to predict effects of DFB exposure on population-level endpoints such as long-term abundance and age structure, and to assess the risk of population decline below threshold abundances.
in a freshwater environment may be overestimating toxicity of effluents to the freshwater compartment.

WE210 Bioluminescent assays as tools for studying antioxidant activity and toxicity of bioactive compounds

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This study promotes application of the bacteria-based and enzyme-based bioluminescent assays to evaluate the antioxidant activity of bioactive compounds in oxidizers solutions. Artificial and natural carbon nanostructures – fullerene derivatives CNM (C60H,Fn) and humic substances (HS) are used here as bioactive compounds. Fullerenols are polyhydroxylated water-soluble derivatives of fullerenes, HS are products of natural transformation of organic matter in soil. Bioluminescent assays allow studying toxicity and antioxidant properties of substances. We studied the toxicity effect of organic (1,4-benzoquinone) and inorganic (K2[Fe(CN)6]) oxidizers on bioluminescence tests. We found the effective concentrations (EC50) of these oxidizers decreasing bioluminescence intensity by 50%. The EC50 values of 1,4-benzoquinone were 2.5±0.1 M and 10 M for bacterial and enzymatic assays, respectively, while the EC50 values of K2[Fe(CN)6] were 4.1±0.4 M and 2.1±0.4 M. Also we studied the influence of bioactive compounds on the assays. They suppressed bioluminescence of the bacterial and enzymatic systems at concentrations >10 M and >5 g/L, respectively. Detoxyfication coefficients can be calculated to characterize changes in toxicity under the action of bioactive compounds. The values of coefficients >1 and So, the bacteria- and enzyme-based assays showed similar peculiarities of detoxification of oxidizer solutions by bioactive compounds. Results show, that low concentrations of bioactive compounds were active. Our work demonstrates a high potential of the bioluminescent assay systems, bacterial and enzymatic, to characterize and compare antioxidant activity of physiologically active compounds.

WE211 Effect of low-dose gamma-radiation on luminous marine bacteria Photobacterium phosphoreum

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The study addresses biological effects of low-dose gamma-radiation. Radioactive 137Cs-containing particles were used as model sources of gamma-radiation. Luminous marine bacterium Photobacterium phosphoreum was used as a biosensor to study the bioluminescent intensity as the physiological parameter tested. To investigate the sensitivity of the bacteria to the low-dose gamma-radiation exposure, the irradiation conditions were varied as follows: bioluminescence intensity was measured at 5, 10, and 20° for 175, 100, and 47h, respectively, at different dose rates (up to 410 mGy/h). There was no noticeable effect of gamma-radiation on fluorescent and DF, while the 20° exposure revealed authentic bioluminescence inhibition. The 20° results of gamma-radiation exposure were compared to those for low-dose alpha- and beta-radiation exposures studied previously under comparable experimental conditions. In contrast to ionizing radiation of alpha and beta types, gamma-emission did not initiate bacterial bioluminescence activation (adaptive response). As with alpha- and beta-radiation, gamma-radiation dependence does not demonstrate monotonic dose-effect dependency. However, the bioluminescence inhibition efficiency was found to be related to the exposure time; while no dose rate dependence was found. The sequence analysis of 16S ribosomal RNA gene did not reveal a mutagenic effect of low-dose gamma-radiation. The exposure time that caused 50% bioluminescence inhibition was suggested as a test parameter for radio toxicity evaluation under conditions of chronic low-dose gamma irradiation. The reported study was funded by Krasnoyarsk Regional Fund of Science according to the participation in the event: «28th Annual Meeting of the European Society for Environmental Toxicology and Chemistry / SETAC Europe 28th Annual Meeting. International» References: [1] Kudryasheva N.S., Petrova A.S., Dementyev D.V., Bondar A.A. 2017. Exposure of luminous marine bacteria to low-dose gamma-radiation. Journal of Environmental Radioactivity 169-170:64-69.

WE212 Bioluminescent Assay for Toxicological Assessment of Nanomaterials

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Due to the increasing scale of production and usage of a vast number of new materials in industrial and economic activities, society is faced with problems associated with a lack of materials safety assessment regarding humans, ecosystems and the biosphere as a whole. Nowadays, numerous toxicological investigations using living organisms, cell lines, etc. are carried out in laboratories in order to assess the potential risks of using these materials and their biological effects on human health and the environment. A rapid bioluminescent enzyme inhibition-based assay was applied to predict the potential toxicity of carbon nanomaterials (CNM) presented by single- and multi-walled nanotubes (SWCNT and MWCNT) and aqueous solutions of hydrate fullerene C60 (C60HyFn). This assay specifically detects the influence of substances on parameters of the soluble or immobilised coupled enzyme system of luminescent bacteria: NADP+/F MN-oxidoreductase + luciferase (Red + Luc). A protocol based on the optical properties of CNM for correcting the results of the bioluminescent assay was developed. If the concentration of the nanomaterial solution was greater than 0.1 in the range of 400-600 nm, the light emission intensity was multiplied by the correction factors. It was shown that the inhibitory activity of CNM on Red + Luc decreased in the following order: MWCNT > SWCNT > C60HyF. The soluble enzyme system Red + Luc had high sensitivity to MWCNT and SWCNT, with values of the inhibition parameter IC50 equal to 0.012 and 0.16 mg/L, respectively. The immobilized enzyme system was less sensitive, equal to C60HyF less than its soluble form, with an IC50 equal to 1.4 mg/L. According to EC Directive 93/67/EEC for aquatic organisms, chemicals are classified by their degree of toxicity based on EC50 values. We hypothesised that this classification was correlated with IC50 values and revealed that MWCNT and SWCNT samples might be characterised as extremely toxic and very toxic, respectively. Due to its technical simplicity, rapid response time and high sensitivity, this bioluminescent method has the potential to be developed as a general enzyme inhibition-based assay for a wide variety of nanomaterials. This study was supported by the Russian Science Foundation (project no. 16-14-10115).

WE213 Delayed chlorophyll fluorescence in biomonitoring of environmental pollution

Y.S. Grigorev, Siberian Federal University / Department of Ecology and Environmental Study; E. Stravinskene, O. Kryuchkova, N. Pakharkova, Siberian Federal University

Plants have important role in biomonitoring of environmental pollution because of their high sensitivity to various pollutants which often disturb photosynthesis. The photosynthetic function of plants is therefore a good indicator to reveal the environmental pollution effects. Fluorescence of chlorophyll is widely used for quick assessment of photosynthesis condition. A prompt fluorescence that can be registered by PAM fluorometers is most commonly used for these purposes. Recently, we have developed a more sensitive and fast indicator of the state of the photosynthetic apparatus of plants, which is based on the measurement of delayed fluorescence (DF) of chlorophyll. Delayed fluorescence is registered in red spectrum area after exciting light flashes in the form of time-attenuated radiation. We found out that the ratio of DF excited by high intensity flashes to DF excited by low intensity flashes decreases more than tenfold when there is a toxic effect on photosynthesis. This indicator can be measured within a few seconds. Due to the relative nature of this indicator, which characterizes the photosynthetic activity of the plant test organism, it does not depend on the size or volume of analyzed sample. To implement this method, we developed the Photon 10 fluorometer which automatically measures the relative indicator of delayed fluorescence (RDF) in 24 plant samples. Simultaneously with RDF, the device registers the variable part of prompt fluorescence of these samples. Applying this device we carried out transplanted lichen indication of air pollution in Krasnoyarsk. We found that the values of DF of several species of lichen differed up to 20% between ecologically clean and industrial areas of the city after three weeks of the exposure. The study of the condition of pine needles in these areas showed a difference in the value of the RDF up to 5 times. Our method also showed a high sensitivity in bioassay of toxicity of natural and waste waters where Chlorella vulgaris algae was used as a test organism. RDF of Chlorella vulgaris absorption decreased by the factor of 2 (EC50) in solutions of copper and zinc ions at a concentration of these heavy metals of 0.005 and 0.5 mg/dm³ respectively.

WE214 Chlorophyll fluorescence temperature curve to estimate changes of the photosynthetic apparatus of coniferous trees during the transition to a state of winter dormancy in urban ecosystems

N. Pakharkova, Y. S. Grigorev, Siberian Federal University / Department of Ecology and Environmental Study; N. Gavsky, Siberian Federal University

The main regulating factor for the transition of plants from active vegetation to winter dormancy is the decrease of air temperature. A gradual decreasing of activity of the photosynthetic apparatus of plants in the period of decreasing air temperature also has a significant influence both during the autumn photoperiodic reaction and at different phases of winter dormancy. This research aims to a better understanding of the responses of the Scots pine and the Siberian spruce to air pollution stress in urban and semi-urban conditions of Southern Siberia. It is well-established that during the transition from the phase of active vegetation into the phase of winter dormancy, the proportion of during the period of dormancy of needles undergo a number of changes. Changes in the assembly of the photosynthetic apparatus are mirrored in changes of fluorescent signals emitted at different temperatures. Chlorophyll fluorescence temperature curve (FTC) is a dependence of chlorophyll fluorescence intensity on linearly increasing temperature. This curve is used for determination of the stability of PS2 and for evaluation of the structural arrangement of chloroplasts in vegetating plants. Also, based on the changes in the shape of the FTC it can be deduced whether the plant is in the state of winter dormancy or it is vegetating. The calculated ratio of the low- and high-temperature peaks (50° and 70°) of zero level fluorescence may be used.
as an indicator of the degree of the depth of dormancy. FTC was measured with the needles’ segments using fluorometer Junior-PAM (Walz, Germany). The needles were linearly heated from 25 to 70 °C at a rate of 2 °C/min using a computer-controlled heating device. In climate conditions of Southern Siberia, disturbance of winter dormancy under air pollution stress represents a major threat to the health status of Pinus sylvestris and Picea obovata. Our data demonstrate that regardless of the age of needles, the degree of winter dormancy of both species clearly correlated with air pollutants, including PM10 and PM2.5. The trees growing in industrial areas are easier to release from dormancy and to be affected by late winter or spring frost. In urban environments the risk of frost injuries is even higher due to early spring warm spells associated with heat island effect. Scots pine is less susceptible to air pollution and temperature fluctuations than Siberian spruce, and therefore represents a better choice for urban forestry projects.

**WE215** Luminessent microscopy in the bioindication of the Baikal pollution with oil products and polyaromatic hydrocarbons

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Against the backdrop of the growing diversity of toxic substances that degrade the natural environment, the creation of new analytical systems for assessing the state of the environment is of paramount importance. Among such systems, fluorescence microscopy is widely used. It has been experimentally revealed that the many pollutants of water bodies, it is necessary to isolate oil products and polyaromatic compounds (PAHs), the flow of which in the valley is constantly increasing. On Lake Baikal, this is due to the development of tourism, the increase in the number of passenger ships and tourist bases, often not equipped with treatment facilities. The necessary system for monitoring the quality of the aquatic environment, including methods for bioindication, not only on generally accepted test facilities, but also on representative hydrobionts for this reservoir. Crustaceans of the order Copepoda, which have well-expressed fatty inclusions in the form of drops, there are several hundred species. Epischura baikalensis Sars (Copepoda, Crustacea) - endemic of Lake Baikal - dwells practically in the entire water column of the pelagic lake. E.baiaclesis accounts for up to 70% of the total biomass of zooplankton. Crustaceans Copepoda, having fat inclusions, accumulate in them oil products. This can, in particular, be observed by the blue-violet glow in a luminescence microscope. The accumulation of diesel fuel by fatty inclusions of Copepoda crustaceans and the high sensitivity of luminescence, makes it possible to assess the concentration of this pollutant in the water under investigation. It is noted that in the presence of unicellular algae that are absorbed by the crustaceans, this process is activated and the accumulation time of oil products and PAHs decreases. Another representative of the Copepoda Cyclops Colensis also has chaotically scattered fatty inclusions in which the accumulation of oil products can be seen in a fluorescent microscope. It has been experimentally revealed that if E. baikalensis with oil products or PAHs accumulated in fatty inclusions is added to pure water to C. Colensis, then the staining of fat inclusions also occurs in the latter, that is, this process is transmitted along the trophic chain. Based on these experiments, a method of bioindication on the accumulation of oil products and PAHs in *Copepoda* crustaceans in fat drops was proposed.

**WE216** The correlation between fluorescent properties of water extract from soil and its effect on bioluminescent enzymatic bioassay

E. Nemtsева, O. Chmurina, Siberian Federal University / Laboratory of Bioluminescent Biotechnologies; M. Gerasimova, Siberian Federal University / School of Engineering Physics and Radio Electronics; V. Kratasyuk, Siberian Federal University / Biophysical

The work is devoted to the development of the bioluminescent enzymatic bioassay of the soil contamination. The problem of relation of bioassay results with intrinsic pollution and the creation of new analytical systems for assessing the state of the environment is of paramount importance. The comparison of enzyme systems for soil contamination bioassay. The study aimed at assessing of the correlation between the content, physico-chemical parameters of the soil solutions including amount of detected arsenic. The conclusion was derived about relation of bioassay signal from studied extracts with the amount of humic substances in soils. The research was supported by the Russian Science Foundation (project no. 16-14-10115).

**WE217** The comparison of enzyme systems for soil contamination bioassay

E. Kolosova, Siberian Federal University / Biophysical; D. Gulnov, Siberian Federal University; N. Rimatskaia, Siberian Federal University / Biophysical; A. Listtis, O. Sutormin, V. Kratasyuk, Siberian Federal University

The work is devoted to the development of the bioluminescent enzymatic bioassay. It has been experimentally revealed that the spectral characteristics of the various soil samples and the results of their bioluminescent bioassay as well as their physico-chemical characteristics. Water extracts from 56 soils (medium and heavy loams, with humus content 1.2-11.3% and detected arsenic 3.05–15.39 mg/kg) were studied by the method of excitation-emission matrix (EEm) fluorescence spectroscopy. The luminescence in the spectral range 290-600 nm under excitation at 250-500 nm was recorded for each extract as well as absorption spectra in the range 200-800 nm. The physico-chemical characteristics of the soils (particle size distributions, pH, humus content, etc.) were compared with the inhibitory effect of water soil extracts on enzymatic bioassay based on the coupled bioluminescent reaction of bacterial luciferase and NAD(P)H:FMN-oxidoreductase. It was revealed that the spectral characteristics of water extracts are similar for all soils and featured by three types of fluorophores with excitation maxima at about 270, 330 and 360 nm and emission maxima at about 330, 425 and 470 nm, respectively. The residual activity of the bioluminescent bioassay enzymes in the presence of soil extracts was found to correlate with intensity of two first bands that is the measure of the component content. Poor correlation was found between EEm characteristics and remaining chemical parameters of the soils including amount of detected arsenic. The conclusion was derived about relation of bioassay signal from studied extracts with the amount of humic substances in soils. The research was supported by the Russian Science Foundation (project no. 16-14-10115).

**WE218** Are changes in bioluminescence kinetics of Photobacterium phosphoreum *P. phosphoreum* to low-dose radiation connected with genetic mutations?

O. Gaseynov, V. Gaseynova, Siberian Federal University; T. Rozhko, Krasnoyarsk State Medical University. prof. VF Voyno-Yasensky; A. Bondar, Institute of Chemical Biology and Fundamental Medicine SB RAS; N. Kudryasheva, Institute of Biophysics SB RAS

Luminous bacteria of marine origin are widely employed as biological sensors for monitoring the environmental conditions and for monitoring the impact of radioactivity. Due to their use as indicator organisms, their use in environmental monitoring and their use in the biotechnology of water purification, bioluminescent bacteria can be used for the detection of biologically active substances in water and soil samples. The study was supported by a grant from the Russian Science Foundation (project No. 16-14-10115).

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**WE220** Are changes in bioluminescence kinetics of Photobacterium phosphoreum *P. phosphoreum* to low-dose radiation connected with genetic mutations?

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**WE221** Are changes in bioluminescence kinetics of Photobacterium phosphoreum *P. phosphoreum* to low-dose radiation connected with genetic mutations?

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not associated with DNA mutations in the gene sequences tested.

LCA and beyond - integrating sustainability and/or other dimensions to improve decision support (P)

WE219 Meet the Framework Regulation and Supply Chain secondary standards in wheat cultivation for sustainable pasta production. An example of broadleaf weed control: halau xenfen-methyl (Arylex™ active) C. Vafi, S. Cavanna, Dow AgroSciences Italia srl; A. Haley, Dow AgroSciences Ltd; W. Jones, DuPont

The need and the awareness of sustainable food production has increased in recent years, driven by the growing awareness of global population increase and its burden on the limited agricultural land available to sustain the required food production. This is being reflected in the choices made at all the steps of food production from field to table. The sustainable production of pasta starts by optimising agricultural practices, which includes the key component of Plant Protection Products (PPP) applied to wheat crops. The development of new Plant Protection Products in Europe is governed by the strictest regulatory framework in the world: Regulation (EC) 1107/2009 concerning the placing of plant protection products on the EU market; Directive 2009/128/EC on Sustainable Use of pesticides and its national implementations (National Action Plans), and Regulation (EC) 396/2005 concerning the Maximum Residue Levels of plant protection products allowed in food. In addition, the Secondary standards coming from Food Processors and Retailers regarding chemical residues in food place increasing standards which have to be considered. The Plant Protection Industry is increasing its focus on sustainable food production not only for complying with the evolving Regulations, but also for helping the farmer to meet the Food Supply Chain needs. A good example of this new emphasis is brought by an innovative auxinic herbicide, halau xenfen-methyl (Arylex™ active), for the control of broadleaf weeds in spring and winter soft and durum wheat. This highly efficacious herbicide requires low use rates and its inherent properties (rapid degradation in the soil and plant) mean a low environmental and human health impact. Utilising halau xenfen-methyl according to its label offers wheat growers a key tool for optimizing production, while producing a commodity with no detectable residues in the grain, in the processed product (flour, bread, wheat germ, malt), and in pasta. Results will be presented and discussed. Therefore, the properties of halau xenfen-methyl are fully aligned with increasingly strict environmental requirements from regulatory authorities and the Food Chain Secondary standards. TM Trademark of Dow AgroSciences

WE220 Cradle to grave Life Cycle Assessment of Traditional and Vegetative roofs J. Koura, University of Balamand / Chemical engineering department; R. Manneh, University of Balamand / Department of Chemical Engineering; V. El Khoury, University of Balamand / Chemical engineering department; E. Andrade, University of Balamand / Chemical engineering department

The aim of this study is to (1) assess the environmental performance of an extensive green roof (EGR) mock-up installed on the rooftop of the Chemical Engineering Department at the University of Balamand, in the region of El Kurah, North Lebanon (34°31′N, 35°50′E) from the raw material phase until the end-of-life phase (through a Life Cycle Assessment (LCA) study); and (2) compare the environmental impacts of an EGR mock-up to a traditional gravel ballasted roof (TGB) mock-up. In this research, the Life Cycle Inventory was modelled using the SimaPro 8.3.0 software and the Ecoinvent database, and the IMPACT 2002+ methodology was selected as the Life Cycle Impact Assessment method. Vegetative roofs seem like a possible solution for the environmental issues in Lebanon since this small Mediterranean country lacks a clear sustainability plan as well as an infrastructure update and only 13.4% of the total surface area (10,452km²) are forested area. Vegetative roofs embellish the unused roof surface available in most urban areas, increase the roof lifetime, reduce the need for a heating/cooling system as a result the building energy consumption is decreased, etc. Vegetative roofs capture a fraction of the rainwater through their growing material and the vegetation could remove airborne pollutants. The findings of this research seem to be very promising, the extensive green roof mock-up has the least environmental impacts for all impact categories except for the “land occupation” impact category due to the vegetation layer. Moreover, a real-time monitoring of temperature was done to assess the electricity consumption or both TGBR and EGR mock-ups within the use phase. Furthermore, the sensitivity and uncertainty analyses will be performed to check the robustness of the results. 

WE221 Filling whole building life cycle assessment gaps for conceptual building design V. Halmo, University of Pittsburgh; J. Chhabra, G. Warn, Pennsylvania State University; M. Bilec, University of Pittsburgh / Civil and Environmental Engineering

Resource consumption, harmful emissions, climate change, and hazard events have triggered increased interest in sustainable and resilient buildings over the past 20 years. The sustainability and resilience performance of buildings has been covered in numerous rating systems and building codes; however, these are typically prescriptive methods focused on setting minimum performance requirements rather than helping us understand and optimize buildings. Life cycle assessment (LCA) and performance-based methods (e.g., energy modeling, seismic loss assessment) can be more beneficial from this standpoint but can be complex and isolated from each other. Most building LCA studies to date have limited their scope to embodied and operational energy use, due to their overwhelming impacts in conventional buildings, while some aspects, such as repair from damages (e.g., seismic) and water use have been largely missing. This study uses LCA, energy modeling, water modeling, and seismic loss assessment simultaneously to obtain a comprehensive view of the costs and benefits of implementing different building design alternatives. Preliminary analysis of a hypothetical building shows that in some metrics, such as cost and eutrophication, repair and water phases, respectively, can contribute more than 10% over the lifetime of the building. One design parameter expected to influence all of the mentioned performance aspects is the building form-factor, and its effect on the results will be presented. Based on the typical approach to building LCA and the shift in the sources of impacts away from operational energy as buildings transition to net-zero energy, this study investigates the potential contribution from missing phases in building LCA. This research brings together knowledge from multiple disciplines and links them through life cycle thinking, investigating the effects of building design decisions in multiple metrics. This approach can be crucial in identifying optimal solutions early on during the conceptual building design phase. 

WE222 Prospects for multidimensional assessment of sustainability in urban environments F. García-González, L. Liñé, Universidad de Santiago de Compostela / Chemical Engineering; A. Núñez, L. González Louro, PEGAMP - Santiago de Compostela; E. Andrade, Universidade de Santiago de Compostela; M. Moreira, G. Feijoo, University of Santiago de Compostela / Chemical Engineering; S. González-García, University of Santiago de Compostela CIF Q1518001A / Chemical Engineering

Urban systems can be considered as living organisms driven by materials and energy flows (urban metabolism). Once these flows are computed, the environmental profile of the city can be analysed. However, when only assessing the environmental aspects of cities, a limited view of their performance is possible, as they are complex systems in which social and economic aspects are at least as important as environmental ones. This fact raises a dilemma, since today’s developed society bases its social and economic wellbeing on the consumption of resources and, therefore, on causing a large impact on the environment. The definition of sustainability includes three main pillars: society, economy and environment. However, considering the social and economic aspects of urban metabolism, two main gaps emerge. The first one is the lack of standardisation. Indicators are used to evaluate these aspects, but different specialized organisms propose different sets of indicators. The second gap is the difficulty of comparing indicators to each other in order to evaluate which city is the most sustainable in a sample. This study aims to fill both gaps by systematically assess the sustainability of several cities in Galicia (NW Spain). To select an appropriate set of indicators, including environmental, social and economic criteria, a Leopold matrix has been constructed considering: i) the data available for the studied case study, ii) the frequency of occurrence in the data sets of specialized agencies (United Nations, European Commission, OECD and The World Bank) and iii) the relevance for the case study. The selected indicators do not have a significant common unit of measurement; therefore, to obtain a common scale for comparisons, all indicators should be normalized. In this study, this has been done by considering unsustainable and sustainable values as reference (Phillis et al., 2017). Finally, a composite indicator, i. e. a sustainability index, is obtained for each city based on the three composite sub-indicators of the sustainability dimensions (environmental, social and economic criteria). This work was financially supported by the Spanish Ministry of Economy and Competitiveness (project ref. CTQ2016-75136-P) and by Xunta de Galicia (project ref. ED431F16/001001(1)). S. González-García would like to express her gratitude to the Spanish Ministry of Economy and Competitiveness for financial support (Grant reference RYC-2014-14984). 

WE223 Life Cycle Analysis of remediation solutions in railways and surrounding areas M. Riera, Leitat Technological Center

An important environmental problem is the pollution associated with trains on external or underground railways. Despite is an issue less studied, a real contamination by heavy metals and hydrocarbons exists in railways, affecting also the surrounding areas. Following the strategic line of Horizon 2020, RECOVER project aims to develop new technologies to reduce the environmental impact of transport systems. The project is executed by COMSA, CETIM and LEITAT, and is within the framework of the call "Challenges of Collaboration" in 2015, with partial funding from the Ministry of Economy and Competitiveness of Spain. The
main aim of the project is to develop systems for the collection and elimination of pollutants (hydrocarbons and heavy metals) present in the ballast and on the ground adjacent to the railway tracks through the use of different technologies: Ballast modified by sol-gel coating based on silicon oxide to capture heavy metals and titanium oxide for the degradation of hydrocarbons. Phytoremediation processes (use of plants to decontaminate soils) and bioremediation (bioaugmentation of the microbial population of the soil) for the uptake of heavy metals and hydrocarbons in the soil adjacent to the roads. The solutions are first performed at the laboratory scale, and subsequently they are located in a real area to evaluate their effectiveness. The remediation procedures are assessed through a comprehensive Life Cycle Assessment (LCA) to identify the environmental benefits obtained with the introduction of the solutions in railways. The environmental analysis includes the life cycle stages of raw materials, accidents, and end-of-life scenarios, including transport and other related aspects. The LCA is performed based on the methodology which is standardized by the ISO 14040 and ISO 14044. Calculations are done using the SIMAPRO software and taking as a basis the Ecoinvent3.1 database. Besides the direct environmental benefits of the pollutants reduction in the soil, the LCA allows the identification of other environmental aspects. The impact categories include microclimate, ozone depletion, photochemical ozone formation, acidification, terrestrial and freshwater eutrophication, and freshwater ecotoxicity. The pollution due to rail transport is a problem identified in Member state of the European Union, the solutions proposed in RECOVERY project could an important contribution to the current railway legislations.

**WE224 Life Cycle Assessment of Asphalt Mixtures vs Road Pavements**
D. Lo Presti, The University of Nottingham / Nottingham Transportation Engineering Centre NTfEC, A. Jimenez del Barco Carrion, The University of Nottingham

Road infrastructures are one of the major assets all over the world. The appropriate construction and maintenance of roads promote economic growth and development of countries. Within the field of road infrastructures, road pavements construction and maintenance require particularly high energy and raw materials consumption and generate elevated GHG emissions. For this reason, great efforts are being made in the last decades to raw materials and energy efficiency of road pavements, including asphalt mixtures. LCA of road pavements is a complex process which needs the collaboration of the different partners involved, such as road authorities or contractors, to provide information, and therefore there exists a common tendency to simplify the LCA of pavements and individually analyse their components - usually asphalt mixtures. This process is not always clarified and may lead to erroneous conclusions or incoherent procedures. In order to use LCA for decision-making in asset management, it must be more related to road pavements rather than asphalt mixtures. In other words, the LCA of the pavement components (e.g. asphalt mixes) should be a mere input and the overall methodology should focus mainly on dealing with data such as road geometry, maintenance strategies, traffic, pavement conditions and statistical parameters to account for data changing over time. The study presented here aims to highlight and quantify the differences between the LCA of asphalt mixtures and road pavements. For this, the different phases of LCA are defined and analysed for the systems respectively. Finally, system boundaries are proposed for each system in order to move towards LCA of road pavements and standardised methodologies.

**WE225 Sustainability assessment of an integrated innovative wastewater and greywater system for an optimal and safe closed water cycle in Mediterranean tourism facilities: demEAUmed solution**
A. Claret, C. Hidalgo, Leitat Technological Center / Sustainability Division; S. Vidal, SANTO TOMAS URBANIZATION CENTER / LEITAT TECHNOLOGICAL CENTER / Sustainability Division

The main objective of demEAUmed is to tackle water scarcity in the Mediterranean area, especially in places with high tourist activity. Also, the project wants to foster the incorporation of sustainability aspects in the tourism sector. To achieve both challenges, demEAUmed has demonstrated the integration of innovative wastewater/greywater treatment technologies to achieve an optimal and safe closed water cycle in Mediterranean tourist facilities. Water resources are limited and unequally distributed geographically and among the year seasons, with higher pressure during summer, in Mediterranean regions. For instance, water consumption per guest has been estimated at 222 L/day in hotels in Spain. So, it is of great importance to achieve a holistic water resource management. demEAUmed affords the reuse of greywater and wastewater generated in tourist facilities with an integrated approach bringing environmental benefits such as water savings and water management carbon footprint reduction. 8 different innovative technologies with an advanced monitoring, control and decision support system have been integrated and implemented on the demonstration site: Samba Hotel-Lloret de Mar, Catalunya, Spain. These technologies have been assessed through a comprehensive LCA, assessing the impacts for each individual technology and for the demo-site integration (7 different configurations). Besides the LCA, a Life Cycle Costing (LCC) is being performed in order to analyse the economic costs. A social LCA (S-LCA) is also conducted in order to assess the social impacts generated by demEAUmed. Life cycle stages of construction and operation of the technologies, main environmental impacts and benefits associated to each technology and combined configurations have achieved important environmental impact savings thanks to the greywater/wastewater recovery and water reuse. As an example, for demEAUmed combined strategies, the carbon footprint is reduced up to 136% (greywater scenario) or up to 62% (wastewater scenario) thanks to water reuse. Focusing on the technologies, main environmental impacts are localized on the operation stage, due to electricity consumption. Concerning the LCC, the overall cost of treating one cubic meter (1m³) of greywater or wastewater by the demEAUmed technologies along their life cycle are being determined. Finally, S-LCA has presented some indicators and the quantification of the socioeconomic impacts and benefits provided by demEAUmed solution.

**WE226 Integrating Life Cycle Assessment and Risk Assessment to support decision making in the framework of Enhanced Landfill Mining**
G. Sarve, KU Leuven Research & Development / Department of Materials Engineering; K. Van Acker, KU Leuven / Materials Engineering

The eco-toxicological impacts would thus be assessed by integrating the environmental impacts of product systems throughout their life cycle. However, when addressing waste management strategies, and in particular landfills, this tool could lead to some limitations. Landfills are in fact highly complex systems and their impacts are affected by several site- and time-dependent parameters. When assessing the potential for enhanced landfill mining (ELFM), the relative perspective of LCA of existing landfill sites and space can become a significant challenge. Integrating LCA and risk assessment to support decision making by defining a consistent “Do Nothing” scenario for landfills for the evaluation of the environmental potential of ELFM. This can be achieved by understanding the processes underlying the emissions of different compounds and estimating the long term emission potential of landfills. In fact, landfill leachate emissions are, on the long term, the major environmental concern and the risks to human health and the environment (HHE) are dependent on environmental and time-dependent conditions. In this context, metal speciation is considered an important aspect to include in the estimation of the emission potential, as the emissions and eco-toxicalogical impacts of metals depend on the variation of site-specific conditions in time. In light of these considerations, a more consistent evaluation of impacts on a global and local scale and considering a long-term perspective could be achieved by integrating LCA with risk assessment (RA), which is a more site-specific tool. In fact, the evaluation of the long-term emission potential of landfills would include the definition of a fate, transport and exposure model for leachate emissions that would then be integrated in the impact assessment stage of LCA. The eco-toxicological impacts would thus be assessed by integrating the variation of pollutants’ concentrations in time and under specific conditions, and by including the variation of background concentrations in the receptor. Literature studies with focus on the integration of spatial differentiation (regionalization) and time-dependency (Dynamic LCA) will be used as references for the study.

**WE227 Comparative environmental sustainability analysis of waste-to-energy techniques for municipal solid waste**
A.J. Ramos, INEGI / INEGI; A.J. Rouboa, University of Pennsylvania / Mechanic Engineering and Applied Mechanics

Progressively advancing societies generate increasingly complex mixtures of residues which led waste thermal treatment methods to evolve greatly in the last decades [1]. Incineration is among the most waste-to-energy (WtE) techniques used for solid residues treatment [2], still gasification is gaining notoriety due to its proven benefits namely concerning efficiency indicators and environmental outputs [3, 4]. Three waste-to-energy techniques for the treatment of municipal solid wastes were considered through a life cycle analysis (LCA): pyrolysis, gasification and combusting. As to make an overall environmental assessment, and to evaluate their environmental impacts: incineration, regular gasification and two-stage plasma gasification. The functional unit chosen was one tonne of residues within similar boundary conditions for each technique. Incineration has shown a sustainable profile, tonnes of debris saving up to 1.3 million kg of resources and materials, while environmental indicators such as global warming potential (GWP), eutrophication potential (ETP) and toxicity potential (TP) depicted enhanced results. Regular gasification uses higher temperatures, thermally decomposing waste and generating syngas which may be utilised as feedstock for further applications. Its environmental impacts revealed poor results for some of the evaluated impact categories, performing worse than incineration. In its turn, two-stage plasma gasification sets an upgrade to more common waste-to-energy techniques, an extra cleaning step to the raw syngas produced by gasification, where even higher temperatures are applied through the use of a plasma torch that literally “melts” the residues. This technique grants environmental benefits such as lower levels of pollutant emissions, less landfilling

WE228 Life Cycle Assessment of Pharmaceutical Waste Disposal in the UK S. Mohamed Yunus, University of York / Environment; A. Boxall, University of York / Environment Department; E. Igos, Luxembourg Institute of Science and Technology / Environmental Research and Innovation

Unused or expired medicines from the hospital and household waste can ultimately end up in landfills or be released to the wastewater system. Therefore, there is the potential for active pharmaceutical ingredients (APIs), from a range of medicinal products, to be present in landfill leachate and sewage effluents. Unused medicines may also be returned to the pharmacist and then be incinerated as hazardous waste. In this project, a household survey was performed to understand the typical waste generation patterns for medicines and the most disposal routes for these substances in the UK. The results show that rubbish disposal (34%) is the common disposal method used by the UK residents with highest estimated emission of APIs to the environment being estimated for paracetamol within the range 7.63 mg/person/day (sewage after sink and toilet disposal) to 76.52 mg/person/day (wastewater after excretion). Based on the survey data, a life cycle assessment study was performed to assess the broader environmental impacts of typical medicinal waste disposal management practices in the UK. The functional unit of this study was 1 tonne of pharmaceutical waste generated in the UK. Three scenarios and treatments explored: incineration after returning back to the pharmacy; landfill treatment after rubbish disposal; and wastewater treatment after toilet and sink disposal. For the life cycle inventory (LCI) phase, two types of databases were used in the study: primary data from the survey study and secondary data from the ecoinvent database. Most of the datasets used in this study are from the whole of Europe (NER) or the United Kingdom (GB). To carry out the life cycle impact assessment (LCIA) phase, the method that has been considered is ILCD 1.0.8 2016 midpoint with APIs. Meanwhile, USETox 2.0 was also used in this study to calculate characterisation factors for the APIs that were not commonly used in the previous study. Furthermore, the impact categories that were considered for this study are climate change, ecosystem quality, human health and resources. The study is still ongoing and the results will be presented at the event.

WE230 Streamlined life cycle assessment of emerging batteries in early design phases using CCaLC tool C. Tomasini Montenegro, KIT, Karlsruhe Institute for Technology; M. Wei, Karlsruhe Institute of Technology KIT / Institute for Technology Assessment and Systems Analysis ITAS

Tomasin-Montenegro, C., Wei, M. M. M. H. HHI, Helmholz-Institute Ulm, Germany b ITAS, Institute for Technology Assessment and Systems Analysis, Karlsruhe, Germany c KIT, Karlsruhe Institute for Technology, P.O. Box 3640, 76021 Karlsruhe, Germany In our modern and globalized society, meeting energy needs in a sustainable way pose one the biggest challenges for the scientific, political and regulatory bodies around the world. Therefore, in the context of the United Nations Development Goals, affordable and clean energy access has been defined as a reachable goal for 2030. In addition to the social impacts associated with this action plan, both tackling climate change and defining regulatory and market frameworks are common elements to identify global solutions for a low carbon energy market. Although it is recognized that geopolitical factors will shape a tailored solution for each geographical region, a transformation of the energy system with a high share in renewable energy sources is necessary to reach a decarbonized energy supply. In particular, considering an energy system with high shares of solar and wind power, energy storage technologies are required to level fluctuating energy production and demand. However, even though when it is recognized that the energy storage technologies exhibit different maturity stages, information about their associated environmental impacts is required to evaluate the sustainability trade-offs inherent to a technology decision-making process. In order to avoid environmental burdens shifting, a life cycle perspective is proposed to develop a model for the preliminary evaluation of emerging batteries or components of these batteries using CCaLC as an assessment tool. The outcome of this work is aimed at contributing to understanding the environmental impacts associated with batteries from a life cycle perspective, while evaluating the advantages and disadvantages of using CCaLC as an assessment tool. 

WE232 Development of Environment Hotspots of Analysis and the consideration of availability to eco-labeling program Y. KURAHARA, N. Isubo, Tokyo City University

In 2014, the UNEP/SETAC life cycle initiative published a guidebook regarding hotspots analysis which enables to extract important elements from the life cycle. They defined this method as “a methodological framework that allows for the rapid assimilation and analysis of a range of information sources, including life cycle based studies, market, and scientific research, expert opinion and stakeholder concerns. The outputs can be used to identify potential solutions and prioritize actions around the most significant environmental, economic, ethical and social sustainability impacts”. Therefore, the scope of hotspots analysis covers environmental and social aspects. The methodology of environmental hotspots analysis has been developed in Japan using latest Japanese inventory database IDEA2 and WIO (Waste Input Output table) and the environmental impact assessment method (LIME). Ideas and applications are evaluated by using this calculation tool. However, the case number of studies using hotspots analysis tool are few, advantages and limitations are unknown. In this research, the usefulness of environmental hotspots analysis through a variety of case studies for Japanese products.

WE233 Environmental burden reduction in the FTA framework using network analysis S. Tokito, Kyushu University

The CO2 emissions embedded in international trade have rapidly increased in countries with lax environmental regulations with expansion of trade and the emission from China and Russia which play a key role in the TPP framework. In addition, with the improvement of environmental efficiency at industry level of a specific country, it is important to corporate within well-specified industrial clusters through supply chain engagement over developing and developed countries (e.g., Kagawa et al., 2015). Moreover, with expansion of trade and the international fragmentation of productions, the promotion of free trade has been increasingly required. In this circumstance, FTA regions have been identified as a significant trade policy issues. For countries participating the FTA framework, it is important to promote economic growth and reduction in various environmental burden simultaneously through the cooperation between the participating countries. To decide the cooperation policy for reducing environmental burden, it needs to understand the structure of complicated supply chain network. However, to the best of our knowledge, previous studies did not analyze the supply chain network induced by the production and consumption of FTA member countries. This study used the network centrality analysis, especially, the applied structural path betweenness (Liang et al., 2016; Hanaka et al., 2017) to EORA database (Lensen et al., 2012, 2013) and analyzed the critical sectors for the cooperation policy in the global supply chain network. From the centrality analysis, a global- and mega-regional” Free Trade Agreement have been identified as a significant trade policy issues. For countries participating the FTA framework, it is important to promote economic growth and reduction in various environmental burden simultaneously through the cooperation between the participating countries. To decide the cooperation policy for reducing environmental burden, it needs to understand the structure of complicated supply chain network. However, to the best of our knowledge, previous studies did not analyze the supply chain network induced by the production and consumption of FTA member countries. This study used the network centrality analysis, especially, the applied structural path betweenness (Liang et al., 2016; Hanaka et al., 2017) to EORA database (Lensen et al., 2012, 2013) and analyzed the critical sectors for the cooperation policy in the global supply chain network. From the centrality analysis, a global- and megaregional” Free Trade Agreement have been identified as a significant trade policy issues. For countries participating the FTA framework, it is important to promote economic growth and reduction in various environmental burden simultaneously through the cooperation between the participating countries.

WE234 Developing life cycle assessment to fight climate change P. Goglio, Cranfield University / School of Water, Energy and Environment; A.G. Williams, N. Balta-Ozkun, N. Harris, Cranfield University / School of Water, Energy and Environment; P. Williamson, University of East Anglia / School of Environmental Sciences; A. Marvuglia, Luxembourg Institute of Science and Technology (LIST) / Environmental Research and Innovation (ERIN); P. Baustert, Luxembourg Institute of Science and Technology LIST / Resource Centre for Environmental Technologies (CRTIE)

Climate change targets could only be achieved with the contribution of greenhouse gases. A large number of greenhouse gases are emitted from several GGR technologies. This study introduces a model to assess the direct carbon capture and storage (CCS), the use of wet raw materials in cement production, enhance weathering, enhancing soil C; forest management; bioenergy and innovation, Chemical and Non-Metallic Mineral Products” in the sector level and “CHN—JPN” in the country level. We can see the large CO2 emission from China and Russia which play a key role in the TPP framework. From the centrality analysis, a global- and mega-regional” Free Trade Agreement have been identified as the most important trade policy issues. For countries participating the FTA framework, it is important to promote economic growth and reduction in various environmental burden simultaneously through the cooperation between the participating countries.

WE235 Development of Environment Hotspots of Analysis and the consideration of availability to eco-labeling program
models in consequential LCA. None of the approaches presented resulted fully comprehensive, with limited uncertainty and complexity. Both approaches based on agent based modelling require a large amount of data and expertise to be used, not often available to the LCA practitioner. The weighted consequential approach has a level of subjectivity higher than other approaches. Instead, the scenario based approach using IAM has a medium level of completeness, uncertainty and complexity. However, the strong basis for IAM is that the model agreement with the IAM assumptions to be used and this constitutes a major limit. The choice among the approaches depends on the objectives of the LCA and should be as complete and comprehensible as possible when climate change claims are made. Future perspectives include a comparative testing of these approaches for selected GGRT and future research should develop and assess potential alternative approaches to those presented. Further research is necessary to develop the appropriate LCA methodology for GGRT.

WE235 HYBRID FULFILMENT-IMPORTANCE MATRIX FOR ASSESSING SOCIOECONOMIC IMPACT

I. Espin Gallart, Fundacio CTM, Centre Tecnologic; J. Berzosa, L. Vendrell, Fundacio CTM Centre; F. Clares, Fundacio CTM Centre Tecnologic. More often, methodologies to assess socioeconomic impact are focused just on determining just a few indicators instead impacts, which don’t use the whole spectrum of socioeconomic insights. In this sense, the hybrid fulfilment-importance matrix emerges with the aim to solve these limitations and to calculate the socio-economic impacts of a new process, service or project. This methodology is not only based in technical performance of the studied system, but also in the holistic approach offered by the LCA, LCC and sLCA methodologies. This semi-quantitative system is based on the scores of the relationship between indicators and impacts. In this way, this methodology allows calculating in which degree the objectives have been achieved, and how the impacts and indicators affect the system. One of the main strong points is its integrated approach which allows to consider the impacts of the project during different dimensions of the project. The indicators, placed in rows, are organized in four categories: technical indicators, environmental indicators, economic indicators, and social indicators. For technical indicators, data coming from performance of the system is implemented. The indicators for the environmental category are taken from LCA studied impact categories, which are supposed to be relevant for the project. In the case of the economic category, indicators studied in the LCC study are considered. Regarding the social indicators, those listed come from sLCA study. The distribution of the columns shows two parts: the fulfilment part, and the relevance part. In the fulfilment part, three columns are deployed: Baseline status, expected results set according to the goals of the new process, service or project, and current or final results. On the importance part, the added columns represent the impacts of the project, which entail technical, sustainability, economic and social insights. When the impacts are selected, its importance in reference to the studied indicators must be defined based on expert know how and opinion. This importance is set by applying a value between 0 and 3. The socio-economic scores are calculated combining the importance values with the fulfilment indicators. As a case study, this methodology has been applied to LIFE RELEACH project, which is aimed at managing leachates coming from landfill by concentrating technologies. In this way, the methodology has allowed to determine which socio-economic impacts have the higher contribution.

WE236 SETAC Sustainability Interest Group

D.L. Carr, Texas Tech University / Biological Sciences

WE237 SETAC LCA Interest Group (Europe)

H. Stichnoth, Thünen Institute / Agricultural Technology

WE238 Life cycle assessment of a thermoplastic starch obtained from mango kernel


Agrifood industry generates large amounts of residues with potential to be used as feedstock, for bio-based products. Mango agroindustry in Brazil is expected to increase to 1.4 million tons by 2024, and processing residues can account for more than 40 %. The mango pulp is the main product, and mango kernel, a so-called residue, is disposed of at landfill, but containing starch, oil and phenolic substances. This study assesses the environmental life cycle impacts of thermoplastic starch produced from mango kernel (MK-TPS), and compare it with fossil-based low-density polyethylene (LDPE). The system boundaries of the MK-TPS start with transportation of mango kernel residue, followed by extraction of starch from mango kernel (together with oil and phenolic substances), and production of thermoplastic. The functional unit adopted was 1 kg of thermoplastic.

A life cycle inventory for MK-TPS was implemented based on primary data gathered at a laboratory scale. Six environmental impact categories were assessed based on the ReCipe life cycle impact assessment method. A sensitivity analysis to the allocation approach for the starch extraction process will be performed comparing mass allocation (56 % starch, 28 % phenolic compounds and 16 % oil) with economic allocation (using a range of expected market prices). Impacts based on allocation for MK-TPS show lower climate change, fossil depletion and ozone depletion, but higher impacts on terrestrial ecotoxicity, freshwater eutrophication and marine eutrophication, comparatively to LDPE. The most important contributor to impacts is starch extraction (due to hexane and methanol), except for marine eutrophication, for which the main contributor is glycerin used to produce the thermoplastic. The paper may contribute to the eco-design of a new bio-based product using a large fraction of the Klingenberg agroindustry as feedstock, through a Life cycle assessment based on laboratory scale data. Future studies shall take into consideration critical aspects and improvement opportunities identified through the study on a larger scale extraction process.

Environmental monitoring of contaminants using terrestrial ecological biomonitor (P)

WE239 Geostatistically estimating spatial structures of heavy metals and nitrogen accumulation in mosses sampled between 1990 and 2015 through Germany and in a bioindicator for atmospheric deposition in Germany. Ecological Indicators, 76:194-206, 2017


Nützen europäischer Messmethoden zur Bestimmung und Regionalisierung von Schadstoffeinträgen für eine Abschätzung des atmosphärischen Beitrags zu land- und forstwirtschaftlichen Anzucht. Acknowledgements: The authors are grateful to the German Environment Agency for funding.

WE240 Semi-volatile organic contaminants (SVOCs) in pine needles from Iceland

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Iceland is famous for a great number of things, but vegetation (particularly forestry) is not one of them. However, trees do exist in this country and pine stands and researches are widespread across the country. The aim of this study was to conduct a biomonitoring of compounds such as the semi-volatile organic contaminants (SVOCs). The most common species are somewhat different from those used more frequently in these kind of studies, but offer nevertheless the same performance and possibilities. Pinus contorta, Pinus mugo, Pinus sylvestris and Pinus cembra needles were collected upon availability in 24 sampling sites that included remote and rural areas but also some urban settlements like Reykjavík or Selfoss. In seven exceptions, heavy metals (since 1990), nitrogen (since 2005) and persistent organic pollutants (since 2010) were determined. Germany participated in all surveys with the exception of that in 2010. In this study, the spatial structures of element concentrations in moss collected between 1990 and 2015 in Germany were comparatively investigated by use of Moran’s I statistics and Variogram Analysis and mapped by use of Kriging interpolation. This is the preconditions to spatially join the moss survey data with data collected at other locations within different environmental networks. The case examples reveal a clear and statistical-significant decrease of concentrations of most heavy metals in moss but not for nitrogen. Due to decreasing element concentrations and the unchanged application of the element concentration classification for the mapping, the heavy metals maps for the survey 2015 do not any longer depict much spatial variation. Therefore, in an upcoming study, this analysis needs to be complemented for the heavy metals by mapping percentile statistics for the whole period 1990-2015 with maps depicting the spatial structure of survey-specific percentile statistics 1990, 1995, 2000, 2005, and 2015.


Acknowledgement - The authors are grateful to the German Environment Agency for funding.
The use of lichens as biomonitors of air quality is inexpensive and effective. We show that for these collections, a few target areas (biological mercury hotspot Kejimkujik and model these regional trends. While broad spa

(we) other than mercury may also help elucidate the potential sources of these elements: Trace metals in lichens can be effectively biomonitored through lichens. Trace metals levels and biological responses investigated and with the distance from the nearest source. A comprehensive study of the lichens in the atmosphere, lake waters and fish is presented in the work. The six remote high mountain lakes investigated were located in the National Park of Aigüestortes i Estany de Sant Maurici (Pyrenees). They encompassed an altitudinal gradient from 1600m to 2500m asl. Active and passive monitoring devices were used for air and water sampling in order to get insight on the presence and environmental fate of organic contaminants over an extended time period. Moreover, brown trout (Salmo trutta) specimens were captured in each lake for bio-monitoring. They were analyzed for contaminants in the muscle, hepatopancreas and gallbladder tissue. The transcriptomic and proteomic analysis of mRNA was also performed to link the levels of pollutants found in these remote high mountains to the effects in these organisms. The observed concentrations will also be compared to past measurements in other high mountain environments for assessment of temporal trends of this background contamination. S. Jarque et al. (2015). Background fish feminization effects in European remote sites. Sci. Rep., 5, 11292.

WE242 Spatial distribution of mercury and trace metals in epiphytic lichens in Nova Scotia, Canada S. Klapstein, Acadia University / Earth & Environmental Science; I. Carvalho, Técnico Lisboa; R. Cameron, Nova Scotia Provincial Government / Department of Environment, where they... Measuring and, depending on the physical-chemical properties of the pollutants, they may bioaccumulate. A comprehensive study of the lichens in the atmosphere, lake waters and fish is presented in the work. The six remote high mountain lakes investigated were located in the National Park of Aigüestortes i Estany de Sant Maurici (Pyrenees). They encompassed an altitudinal gradient from 1600m to 2500m asl. Active and passive monitoring devices were used for air and water sampling in order to get insight on the presence and environmental fate of organic contaminants over an extended time period. Moreover, brown trout (Salmo trutta) specimens were captured in each lake for bio-monitoring. They were analyzed for contaminants in the muscle, hepatopancreas and gallbladder tissue. The transcriptomic and proteomic analysis of mRNA was also performed to link the levels of pollutants found in these remote high mountains to the effects in these organisms. The observed concentrations will also be compared to past measurements in other high mountain environments for assessment of temporal trends of this background contamination. S. Jarque et al. (2015). Background fish feminization effects in European remote sites. Sci. Rep., 5, 11292.

WE243 Biological monitoring of environmental quality near a solid waste incinerator in central Lithuania G. Sujetoiene, P. Smailigatis, Vytautas Magnus University Waste disposal has huge environmental impacts including toxins, leachate and greenhouse gases. Lichen E. farinacea was transplanted from one old (L.) Ach. were used for biomonitoring the effect of one of the largest solid waste landfill in central Lithuania. Lichen transplants were exposed for 3 months. Chlorophyll content increased in both transplanted lichens with increase in distance from the landfill. Chlorophyll content in lichens was significantly lower in the nearest study site in comparison with the control. Potential quantum yield expressed as Fv/Fm, in thalli was lower under the influence of solid waste incinerator in comparison with the reference. Higher chlorophyll degradation was characteristic to the transplanted lichens under the influence of landfill. The conductivity of leachate and content of thiobarbituric acid reactive substances (TBARS) increased in lichen material transplanted at sites facing the landfill. The results showed that biological monitoring can be useful tool for landfill monitoring.

WE244 What is it means: Levels of PCDD/Fs in the surroundings of a hazardous waste incinerator M. Marques, Rovira i Virgili University / Chemical Engineering; M. Mari, Universitat Rovira i Virgili / Chemical Engineering; M. Nadal, Universitat Rovira i Virgili / School of Medicine, ISEP; M. Schuhmacher, Rovira i Virgili University / Department d Enginyeria Quimica; J. L. Domingo, Universitat Rovira i Virgili / Laboratory of Toxicology and Environmental Health Soil and vegetation were used as environmental monitors to assess the occurrence of dibenzofurans (PCDD/Fs) in the vicinity of a hazardous waste incinerator (HWI) located in Tarragona (Spain). Results belonging to 2015 and 2016 were compared to a previous study conducted in 1998, before the plant started operating, to evaluate the potential impact of the facility after several years of regular operation. The median concentrations of PCDD/Fs in soil samples collected around the HWI were 0.46 pg I-TEQ/g (dw) (range: 0.14 to 1.96 pg I-TEQ/g (dw)) and 0.44 pg I-TEQ/g (dw) (range: 0.13 to 1.34 pg I-TEQ/g (dw)) in 2015 and 2016, respectively. No statistical differences were found between 2015 and 2016 campaigns. Comparing the study of 1998 (median: 0.75 pg I-TEQ/g (dw)) with these carried out in 2015 and 2016, the concentration of PCDD/Fs statistically decreased by 41 and 55%, respectively. Median concentrations of PCDD/Fs in samples of vegetation collected in the vicinity of the incinerator were 0.25 pg I-TEQ/g (dw) (range: 0.11 to 0.68 pg I-TEQ/g (dw)) in 2015 and 0.17 pg I-TEQ/g (dw) (range: 0.09 to 0.36 pg I-TEQ/g (dw)) in 2016. The temporal trend of PCDD/Fs in vegetation was very similar to that of soil, with overall reductions of 4%, 30% and 27% over the period 1998-2015, 1998-2016 and 2015-2016, respectively, being statistically significant in the two latter periods. Although the concentrations of PCDD/Fs in both soil and vegetation samples collected in urban areas showed higher levels than those from rural areas, there was no direct relationship between the levels of PCDD/Fs and the distance or proximity to the plant. In addition, the comparison of PCDD/Fs profile of chimney emissions and the corresponding samples of soil and vegetation denotes noteworthy differences in the contribution of some congeners. Consequently, there is a low potential impact of the plant on the environment, regarding to the emission of PCDD/Fs. Finally, concentrations of PCDD/Fs in soils and vegetation here reported are similar and/or below those observed in the scientific literature for similar areas.

WE245 The use of land snail Cornu aspersum as sentinel organism to monitor air pollution L. Sturb, M. Vannuccini, G. Liberati, F. Nannoni, G. Protano, University of Siena / Department of Physical, Earth and Environmental Sciences; N. Fattorini, University of Siena / Department of Life Sciences; I. Corsi, University of Siena / Physical, Earth and Environmental Sciences The use of biotickorganisms for monitoring air pollution allow to assess real hazardous effects of airborne contamination over a geographical and temporal scale depending on selected species and scientific approach. The present study aimed to validate the use of the land snail Cornu aspersum as bioindicator of airborne pollutants by transplanting snails in plastic cages positioned in urban area strongly impacted by several industrial activities nearby. Ten sites were selected based on their geographical directions from the main industrial area in order to assess pollutant distribution in terms of bioavailability and biological responses in a relatively short period of time. After four weeks trace metals levels in soft tissues of whole organism and several biochemical responses were investigated in different tissue and organs such as: lysosomal membrane stability (LMS) and Micronuclei (MN) in hemocytes and antioxidant enzymes Catalase (CAT) and Glutathione-S-transferase (GST), lipid peroxidation (MDA) and total Metallothionein proteins content (MTs) in midgut. Results obtained by generalized linear mixed models (GLMMs) revealed significant correlations among trace metals levels and biological responses investigated and with the distance from...
the major industrial site. Based on such findings and previous evidences of the ability of this species to respond to vaporized metals as cadmium in laboratory controlled condition, the present study support the suitability of C. aspersum as bioindicator for heavy metals exposure in air pollution monitoring studies.

WE246

The relationship between lead exposure on dogs and their behavior around Pb mining area, Kabwe, Zambia

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Lead (Pb) toxicity on both of human and animals has been known and caused neurological symptoms and even death in the worst cases. Our previous study has revealed Pb exposure on domestic dogs around a Pb mining area, Kabwe, Zambia. There was a trend that Blood Lead Levels (BLLs) in dogs in sites near to the mining area were higher than those in sites far from the mining area. Moreover, the difference of BLLs in the same area among individuals was found. This difference may be attributed to some factors, and behavior of dogs could be one of the important factors. The present study was undertaken to determine a relationship between Pb exposure on domestic dogs and their behavior recorded using GPS machines around the mining area in Kabwe, Zambia. Blood samples of domestic dogs which were freely roaming in the area were collected twice before putting GPS and after a week. BLLs were analyzed by LeadCare II. GPS devices were set to log every 1 minute and 30 seconds and attached to dog collars. In total, 53 male and 48 female domestic dogs were sampled. The overall mean of BLLs before and after a week were 24.2 μg/dL and 24.8 μg/dL, respectively. There was no significant difference between BLLs before and after a week. GPS log data was averaged collected for 4.4 days and the means of distances of dogs movements per day were 17.6 km. There was no significant relationship between distances of dog movements per day and the gap of BLLs in dogs between before and after a week. The distance between the mining area and dogs’ home was significantly negatively correlated with BLLs (p < 0.05). The previous study revealed the concentrations of Pb in soils were negatively correlated with distance from the mine. These trends suggest that the distance from the mine is a key factor of Pb exposure on dogs. In the present study, there was a significantly negative correlation between BLL and distance between the mine and the dog’s home. In contrast to our hypothesis, there was no significant relationship between BLLs in dogs and their behavior. It suggested that mature dogs in the study area could be highly exposed to Pb regardless their behavior. An additional study focusing on the behavior of young dogs which are more vulnerable to Pb could get a different result of the relationship with their Pb exposure.

WE247

Monitoring and impact assessment of terrestrial ecosystem using Eisenia fetida affected by chemical incidents

K. Kim, H. Jeon, H. Kim, Y. Kim, Y. Choi, S. Lee, Kyungpook National University

Many chemicals can be accidently spilled in the environment and it is important to know their presumable toxicities on the living organisms to determine risk assessments. There are no information on the terrestrial organisms of six chemicals possibly spilled into the environment, containing sulfuric acid, methanol, methylhexylketone, nitric acid, formic acid, and toluene. In this study, we conducted acute toxicities of these six chemicals on Eisenia fetida in an artificial soil according to the OECD guideline 207. We used E. fetida adults grown in our laboratory for 10 generations in soil consisting of pig manure composts fortified in an artificial site. The earthworms used in this study were sexually well developed with an average body weight of 100 to 200 mg. The distance between the mining area and dogs’ home was significantly negatively correlated with BLLs (p < 0.05). The previous study revealed the concentrations of Pb in soils were negatively correlated with distance from the mine. These trends suggest that the distance from the mine is a key factor of Pb exposure on dogs. In the present study, there was a significantly negative correlation between BLL and distance between the mine and the dog’s home. In contrast to our hypothesis, there was no significant relationship between BLLs in dogs and their behavior. It suggested that mature dogs in the study area could be highly exposed to Pb regardless their behavior. An additional study focusing on the behavior of young dogs which are more vulnerable to Pb could get a different result of the relationship with their Pb exposure.

WE248

Biochemical and behavioural responses in two endogeic earthworm species exposed to parathion

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The earthworm species Eisenia fetida is a common organism in the soil toxicity testing framework, however, recent studies have point out endogeic species are more sensitive to pesticide than E. fetida. Moreover, interspecific differences in the response of this ecological group of earthworms to agrochemicals should be investigated for a better understanding of pesticide impact at population level. Herein, two endogeic and aburanean species in the agroecosystem Anthocoris chlorotoxica and Aporrectodea caliginosa were incubated in Oleabladan® (ethyl parathion)-contaminated soils. Behavioural (burrowing, casting and feeding, this latter assessed through earthworm mass changes) and biochemical (acetylcholinesterase [AChE] and carboxylesterase [CBE] activities) were measured after 7 days of pesticide exposure. Our results clearly showed species-specific differences in sensitivity and bioaccumulation of A. caliginosa and A. chlorotoxica, indicating A. caliginosa is the most sensitive species to this pesticide under the exposure conditions of our study. Although CBE activity was determined in an attempt to account for these interspecific differences because the implication of this esterase activity in organophosphate detoxification, we found that CBE activity of both species had the OPsensitivity. However, an in vitro inhibition trial with ethyl parathion evidenced a higher sensitivity of A. caliginosa AChE activity compared with that of A. chlorotoxica, which suggested that this toxicological endpoint may contribute to the interspecific differences of behavioural responses such as cast production rate. Our findings suggest the use of more than one endogeic earthworm species to assess toxicity from organophosphate insecticides, overall when these earthworms have a beneficial impact on soil fertility.

WE249

Cr transport in sweet peppers plants cultivated with vermicomposted tannery wastes

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Untreated wastewater and solid waste generated by the tanning industry can cause serious environment damage, particularly to surface and groundwater. In order to avoid pollution, specific chemical, physical and biological treatments have been adopted to reduce waste toxicity, causing mainly by the presence of the chromium. Chromium exists in oxidation states of Cr (III) and Cr (VI). As it is well known, the trivalent oxidation state is the most stable form of chromium and it is essential to plants in trace concentrations. In other hand, the hexavalent is toxic and carcinogenic to mammals, even in small concentrations. Thus, the aim of this work was to investigate the Cr transport in sweet peppers cultivated with vermicompost in order to investigate the contamination by exchanging transport from the vermicomposts and its possible transportation through the plant, the content of Cr (III) and Cr (VI) were evaluated in all the binary soil-plant: soil (at digging and post-harvest), roots, stalk, leaves, and mature fruits. Cr (III) and Cr (VI) were determined through graphite furnace atomic absorption spectrometry (GF AAS). Values of Cr (VI) were below the detectable level (LOQ) in all the analysis. In general, all treatments showed a decrease on their Cr (III) content during the sweet pepper cultivation. The concentration of Cr (III) varied in leaves < stalks < roots < fruits. Values of chromium were in accordance with the maximum permitted in the Brazilian legislation for food security; Cr (VI) was not reported in any sample. A significant quantity of Cr (III) decreased between the concentration at the beginning and at final experiments (post-harvest). Assessing the dynamics of the Cr (III), root uptake should be considered. These results followed the order: the empty pot > the vermicompost. Differences in the chromium contents were not observed between samples and treatments which received vermicomposted tannery wastes with others, without addition of chromium residues. Keywords: vermicomposting; tannery wastes; chromium; sweet pepper

WE250

Insecticide resistance in the natural enemy F. auricularia: detoxification pathways and sensitivity of acetylcholinesterase to organophosphosphate insecticide.

A. Le Noyeav, UAPV/IMBE/INRA; M. Siegwart, INRA Avignon / Unité PSH, ENSAT, Université de la Route de l’Intelligence, Site Agrocampus; Y. Capowiez, INRA Avignon; M. Rault, UAPV/IMBE / IMBE UAPV UMR 7263, Pôle Agrosiences. Apple orchards are highly treated crops, in which organophosphorus (OP), neonicotinoid and synthetic pyrethroid compounds were heavily sprayed on target species. These compounds are toxic to non-target arthropods and increase the risk of resistance making apple orchards an interesting case to study the deleterious effects of insecticides on non-target arthropods and the leafhoppers. The detoxification pathway of reduced pesticide use, the development of biocontrol agents has to be promoted. This work focuses on the assessment of the resistance/tolerance to OP insecticide of the earwig Forficula auricularia, an effective generalist predator. The threshold activities of enzymes that belong to detoxification pathways involved in insecticide resistance were estimated depending on the origin of earwigs. Then, variations in those activities were assessed under environmental conditions prior and after exposure to normal application rate of chlorpyrifos. Adult earwigs were sampled in apple orchards conducted under different management strategies: conventional, Integrated Pest Management (IPM), reduced pesticide use thanks to mating.
disruption practice), and organic ones. Two frequently involved in pesticides resistance enzyme families: Glutathione-S-transferases (GST) and Carboxylesterases (CbEs) were studied, by measuring their activities on electron extracts. Acetylcholinesterase (AChE) activity, the molecular target of OP insecticides, was monitored as toxicological endpoint. We observed that the mortality rate of adult earwigs exposed to the authorized dose of chlorpyrifos depends on the concentration of earwigs in the environment. AChE activity appears to be highly inhibited in earwigs from organic or IPM compared to conventional orchards. Moreover AChE inhibition increased when earwigs were exposed to both chlorpyrifos combined to a specific inhibitor of CbEs. Moreover, we observed that basalt-activities of CbEs and GST of unexposed individuals are higher in conventional orchards compared to IPM and organic ones. All these observations support the hypothesis of a molecular target modification in AChE decreasing to a degree of affinity with the insecticide, and highlight the role of CEs ensuring effective protection of AChE. Our findings suggest the acquisition of resistance to chlorpyrifos in earwigs caught in conventional orchards and point out the necessity to understand these mechanisms in order to evaluate their relevance as biocontrol agents.

WE251
Bioaccumulation of persistent halogenated organic pollutants in insects: Common alterations to the pollutant pattern for different insects during metamorphosis
L. Yu, Guangzhou Institute of Geochemistry / State Key Laboratory of Organic Geochemistry; X. Huang, Guangdong Key Laboratory of Environmental Resources Utilization and Protection; X. Luo, Guangzhou Institute of Geochemistry / State Key Lab. Organic Geochem; L. Tao, Guangzhou institute of Geochemistry Chinese Academy of Sciences; Y. Zeng, B. Mai, Guangzhou Institute of Geochemistry Ubiquitous use of halogenated organic pollutants (HOPs), such as polychlorinated biphenyls (PCBs), polybrominated diphenyl ethers (PBDEs), and dichlorodiphenyltrichloroethanes (DDT), can accumulate in organisms and become magnified along the food chain. Insects play an important role in the transformation of pollutants from abiotic to biotic media. However, few studies have been discussed the accumulation and fate of contaminants in insects. Furthermore, metamorphosis effects on the stable isotope signatures and enrichment characteristic of contaminants in insects, but the bioaccumulation pattern and mechanism during metamorphosis is not well understood. Therefore, we detected the concentrations of halogenated organic pollutants in four taxonomic insects (dragonfly, butterfly and moth, grasshopper, and litchi stinkbug), including three kinds of metamorphism type, collected from an e-waste pollution region in South China. Stable isotopic analysis showed grasshopper have the highest δ13C value, indicating a C3-plant-based food source. In contrast, the butterfly, moth, and litchi stinkbug all represent a C4-based diet preference characterized by lower δ13C values. Moreover, enrichment of the heavy N isotope during metamorphosis is observed in the dragonfly and litchi stinkbug, but the other species (grasshopper, butterfly and moth) did not show significant increases in the values of δ15N from larvae to adults. Principal component analysis (PCA) was conducted using the fraction composition of HOPs were performed to evaluate the species-specific bioaccumulation. Different species of insects exhibited different contaminant patterns, which could be attributed to their habitats and feeding strategies. For example PBDEs were predominant in the dragonfly collected from the pond, which has been seriously contaminated by electronic waste; however, DDTs significantly contributed to the total HOPs in the butterfly and moth, and in the litchi stinkbug, and in their host plants also have a high DDTs concentration. In addition, common multi-linear correlations between ln adult/larva and log Kow of the compound was observed for the four taxonomic insects. The ratio of larva to adult decreased with increasing values of log Kow (log Kow = 6-6.5), then increased (6 < log Kow < 8) and decreased again (log Kow > 8). The results of this study demonstrated that a common mechanism is responsible for the fate of HOPs during metamorphosis in those insects.

WE252
Glyphosate: toxic or not toxic, this is the question.
M. Verderame, R. Scudiero, University Federico II / Department of Biology Insect xenobiotic resistant (XR) and biodegradation (BDG) biocatalysts are important enzymes involved in the detoxification process of various xenobiotics in the plant kingdom. Glyphosate (GLY), a broad spectrum herbicide widely used in agricultural, industrial and urban areas, is a great matter of debate. Although classified by the EPA as “non-toxic and not an irritant” and by the EFSA as “non-carcinogenic to humans”, converging evidence suggests that GBHs, such as Roundup (Monsanto), pose serious health risk on the environment, these molecules caused the contamination of both fresh and seawaters, entailing their accumulation in the biota. Seabirds are highly exposed to environmental contamination because of their ecological habits, high trophic position in the marine food chain, and migration routes. Contamination in birds can be transferred to the offspring via their eggs, which are considered as good bioindicators of environmental pollution for a plethora of contaminants. However, the information concerning the maternal transfer of PFAs in bird eggs and their variation according to the laying order is still inadequate. Thus, the aim of the present study was to determine the levels of PFAs in three-egg clutches of the yellow-legged gull (Larus michahellis) breeding in a colony located at the Comacchio lagoon (Northern Italy) and their variation according to the position in the laying sequence. Eleven perfluoralkyl acids (PFAAs) were analyzed in the yolk of eggs sampled at the time of deposition from 15 three-egg clutches. Independently of the laying order, perfluorooctanoic acid (PFOS) was the main compound detected in the egg yolk, followed by perfluorooctanoic acid (PFOA) and perfluorododecanoic acid (PFDoDa). Moreover, the ΣPFs decreased according to the position in the laying sequence, with first- and second-laid eggs showing higher concentrations compared to last-laid eggs. A similar decreasing trend was also noticed for single compounds, namely PFOS, perfluorononanoic acid (PFNA), perfluorodecanoic acid (PFDA), perfluoroheptanoic acid (PFHpA) and PFDoDa, with concentrations measured in the last-laid eggs that were significantly lower compared to those from the first- and second-laid eggs.

WE254
First assessment of metal concentration in the crab Goniopsis cruentata (Lamarck, 1819) (Decapoda, Grapsidae) from two brazilian mangroves areas with different levels of contamination
M. Vedolin, University of São Paulo USP; T.H. Trevizani, Universidade de Sao Paulo / Instituto Oceanográfico; M. Petti, University of São Paulo USP; R.C. Figueira, University of São Paulo USP / Institute of Oceanography The crab Goniopsis cruentata is a common semi-terrestrial species in brazilian mangroves. Its geographical range includes the western Atlantic Ocean from Bermuda to Brazil, and the eastern Atlantic Ocean from Senegal to Angola. The species is an important fishery resource for traditional communities in the some regions of Brazilian coast. These ecosystems are located in regions of intense anthropic activity and have been proved to accumulate heavy metals. The use of congeners of these bioavailable heavy metals (Sn, Pb, Zn) allows to outline comparisons over space and time and provides significant ecotoxicological integrated measures of the selected metals within the studied system. This study aimed to assess the levels of metals (As, Cd, Cu, Cr, Ni, Pb and Zn) in different tissues (muscle, hepatopancreases and gills) of G. cruentata and compare populations from contaminated and non-contaminated areas. Samples were collected in two mangrove areas of different levels of contamination: São Paulo State, close to a highly industrialized area, and a non-contaminated area during a period of one year, to assess the bioaccumulation of metals associated with seasonality. A two-way ANOVA was carried to analyze interactions between season and sites. The results showed high concentrations of metals in the gills, which was considered a strong reflection of high exposure of G. cruentata to these contaminants. In general, the hierarchical pattern of metals concentration in organs was represented by gills> hepatopancreases> muscle, except for Zn. There were significant differences in metal levels between seasons and sites (p<0.05). The highest concentrations were observed in the summer, which corresponds to the period of greatest metabolic activity of the organisms. Surprisingly, organisms from
unpolluted regions, accumulated more metals than from polluted areas. Thus, we conclude that there are external factors (grain size, pH, salinity) that reduce the mobilization of these chemicals to the tissues and, consequently, their bioavailability to the local biota. Therefore, studies of metal concentrations in mangrove areas are relevant and useful for monitoring the health of environment, maintenance of biodiversity, and for assuring the quality of life, mainly for human when consumed.

WE255 Maternal Transfer of persistent halogenated organic pollutants in Watersnakes (Enhydris chinesis) X. Luo, Guangzhou Institute of Geochemistry / State Key Lab. Organic Geochem; L. Wu, Guangzhou Institute of Geochemistry, Chinese Academy of Sciences / State Key Laboratory of Organic Geochemistry; B. Mai, Guangzhou Institute of Geochemistry

Halogenated organic pollutants (HOPs) such as polychlorinated biphenyls (PCBs), polybrominated diphenyl ethers (PBDEs) and dichlorodiphenyltrichloroethanes (DDTs) are ubiquitous contaminants in the environment. Maternal transfer of HOPs to the offspring is a selective process in which the fat content of fish is related to the distribution of contaminants in ovioparous species. Secondly, due to relatively high lipid and weight of egg in watersnakes, the ratios of contaminant burden in egg over the sum in muscle and egg (EMER) was used to evaluate the tissue distribution of contaminants in watersnake eggs. The values of EMR, and EMER were respectively 2.93 and 95% for PCBs and 0.35 and 66% for PBDEs. Meanwhile, DDTs, PCBs, PBBs, HBB, PBB 153 and lower-brominated BDE congeners showed the ratios of EMR, and EMER higher than 1 and 88% (the lipid percentage of egg to egg plus muscle), respectively. The results indicated that these chemicals were readily transferred from muscle to egg or preferential accumulation in egg compared with muscle. Other chemicals, such as higher-brominated BDE congeners, DP, PBB209, and DBDPE, showed ratios of EMR, and EMER lower than 1 and 88%, respectively, indicating less readily maternal transferred or a preference for muscle. A multi-linear relationship exists between EMER and log KOM, the chemical of the watersnake. For compounds with high hydrophobicity (log KOM < 8), a negative relationship between EMER and log KOM is observed (p < 0.05). With the increase of log KOM, the value of EMER increased. Few significant variety (p = 0.19), all greater than 90%. Maternal transfer potential and the deposition of contaminants in watersnake eggs are obviously different with other species in previous study, which implied potentially high-inter-species differences in the maternal transfer mechanism.

WE256 Development of a Multi-compound Multi-matrix Method for Analysis of Halogenated Flame Retardants Comprising a Multi-step Cleanup and Use of GC-API-MS/MS and GC-El-MS F. Neugebauer, Eurofins GfA Lab Service GmbH / R&D; A. Dreyer, Eurofins GfA GmbH; N. Lohmann, Eurofins GfA Lab Service GmbH; J. Koschorreck, Umweltbundesamt

The ultra-trace analysis of halogenated flame retardants (HFR) leads more and more to the question of analyzing substances of actual or future relevance such as e.g. Dichloro-Plus and other dechloranes or novel brominated flame retardants together with legacy compounds as PBDEs. In order to address this issue, comprehensive analytical methods covering at the same time compound groups of different chemo-physical properties are more and more required, especially for monitoring purposes like analyses within environmental specimen banks. The presented method is validated for a broad range of different environmental matrices (spruce shoots as representatives for plant materials, bream fillet as representative for animal tissue, herring gull eggs as representatives for bird eggs and riverine suspended particulate matter as representatives for organic matter rich in solids) and presently capable of analysing 21 alternative HFRs and 24 PBDEs. The analysts cover different chemical substance groups from Dichloran Plus and other dechloranes to brominated benzenes and alkyl benzenes, ethers and esters (TBA, ATE, BATE, PBT, PBB, HBBz, DPTE, BDEPB, EPTBB, BTBPE, Dec602, Dec603, Dec604, DPM, Cl10-antiDP, Cl11-antiDP, syn-DP, anti-DP, DBDPE). In this way, it gives an analytical basis for further extension towards other compounds. Especially regarding different column chromatographic clean-up steps and use of modern analytical equipment as e.g. a GC-API-MS/MS-system, pointing out possibilities and limitations of such a broad scope of analytes.

Product benefits and positive outcomes: valuation and beyond (P)

WE257 A method to calculate carbon footprint T.K. Paulya, S. Vatanen, VTT Technical Research Centre of Finland Ltd; K. Grönnman, R. Soukka, Lapinpuisto University of Technology

Environmental impacts are typically assessed by measuring and modelling the negative effects that products, services and companies cause to the environment. In practice this means evaluating the used resources and energy and the emissions caused. However, many companies do good causing little. Positive environmental impacts. The strong need for communicating the positive environmental impacts has been identified e.g. by Pihlola et al. (2010). Only, we lack systematic methods to quantify and communicate these impacts that are also called handprints. This presentation proposes a concept to assess and communicate the carbon footprint of a product. The method is in line with life cycle assessment (LCA) and footprint methods and is built on the principle that reducing one’s own foot print is not a footprint. Instead, the handprint comes through improvements caused in the performance of another actor. The most fundamental parts of defining the carbon footprint are to recognize the mechanisms of forming the handprint and to determine the baseline. The carbon handprint can be created via more efficient material or energy use, by replacing or avoiding unwanted materials, waste reduction or extended service life and reuse. Also carbon capture and storage is a way to contribute to carbon handprint. The paper demonstrates through case studies situations where different approach for the determination of the baseline is required. The quantification of the carbon handprint requires several carbon footprints calculated in order to find out if the new solution or product actually reduces the carbon footprint of another actor and cycle stage. The footprints are used in a target actor using the baseline solution, the new solution and the target actor using the new solution.

WE258 Assessing regionalised Life Cycle Assessment (LCA) and economic values of ecosystem goods and services: Impacts of upstream natural land transformations on ecosystem quality A. Ajayebi, University of Exeter / Renewable Energy

Setting up operational and spatially-explicit sustainability assessment models with practical levels of data requirement is becoming more essential as the trend of globalising economy is stagnating and accounting for impacts of human activities is becoming more complicated. Here we developed a model based on regionalisation of Life Cycle Assessment (LCA) that is capable of employing a holistic perspective while taking into account natural land transformations that are related to the life cycle processes. Furthermore, our model can interpret the impacts of land transformations on the ecosystem quality. Economic values of Ecosystem Services (ES) are used as an indicator to the difference between the value of land before and after transformations is representing the damages to the ecosystem quality. We performed a case study for the deployment of a 10 MW photovoltaic solar farm in the UK. The results demonstrated that the upstream life cycle processes transform 6354 m² of natural land into artificial land covers. We also estimated these that transforms reduce the ecosystem service value of the transformed natural land in a cycle upstream cycle stage. The processes from $2364/year to $1587/year, resulting in a decrease of $765/year. The majority of ecosystem service value decreases are in China, where the manufacturing processes take place. The rest of Asia, Australia, Russia and Africa also suffer from noticeable ecosystem service value decreases because of upstream production industries. Europe, where the solar farm is deployed, only suffers modest ecosystem service value decreases due to upstream processes. In comparison to other land use life cycle impacts, this implies that despite the apparent advantages of ‘clean’ renewable electricity production at the deployment location, the majority of ecosystem service damages might be relocated to other parts of the world.


At the recent World Forum on Natural Capital (27-28 November 2017) a wide range of corporate and investor leaders gathered to discuss progress on accounting for Earth’s natural capital - the challenges, the innovations and the actions still needed. This poster will bring some personal reflections of the conference, including key findings from related recent literature, and elaborate on how scientists at SETAC Europe might engage with developments in natural capital accounting. Recent publications, such as “Can we stop depleting national capital?” (Cohen and others 2015) highlight the need to develop and global financial prosperity yet scientific research shows that some natural capital is in a poor state, and declining further. The report finds political and economic systems are unprepared for responding to the risk of natural capital degradation for three reasons: (i) natural capital is not being accurately measured or valued in the context of ecological tipping points; (ii) aggregate economic models are ill-equipped for seeing the dependencies between ‘capitals’ as most cost-benefit analyses used in everyday decision analyses assume that natural capital can be easily substituted by manmade capital, when in fact it cannot; and (iii) we lack appropriate political and economic institutions to manage natural capital effectively. Two opportunities emerge
including all natural capital could support greater prosperity if it were more appropriately valued and hence more efficiently used. The second is governance regimes based on scientifically informed political decisions should protect natural capital. Governance of natural capital stocks should be informed by biophysical limits, potential irreversibility, thresholds and risks to essential function. At the global level, the UN Sustainable Development Goals apply to all countries and provide a foundation for such a governance framework. We are all consumers and beneficiaries of natural capital, but it seems clear that SETAC members can also inform the debate. As environmental scientists we can contribute to developing an understanding of criticality – the tipping points for ecosystems, in particular where these relate to chemical pressures. There are opportunities to work with economists to ensure natural capital accounting and valuation is accurate and measurable, preferably quantitative. One of the strongest themes was that we all need to improve our story telling across multiple disciplines and institutions.


The sustainability of hybrid-electric vehicles (HEVs) has been called into question (Bailey et al., 2017, Hickman, 2012, Publishers, 2008, Biello, 2016). With the recent onslaught of HEVs to the motor vehicle market, there is a need for the internalization of the costs – both environmental and economic – associated with one of its most valuable parts, the HEV motor. For this purpose, a screening Life Cycle Assessment (LCA) and Life Cycle Costing (LCC) study has been carried out on the entire value chain of a dismantled HEV motor. The results of this study are analyzed quantitatively. The aim is to identify bottlenecks for such a quantitative analysis as well as to identify the hotspots from both an environmental and economic point of view. Furthermore, the results will serve as one of the important inputs in laying the foundation for the development of the sustainabiliy boundaries of this study are from cradle inventory (inputs, outputs and emissions) per each food product. The system developed to compute total life cycle costs of a HEV motor. The analysis considered capital and operating costs in order to define the total vehicle cost of ownership over 10, 15, and 20 year life expectancies. The development of an integrated life cycle costing and life cycle analysis will allow any individual to evaluate properly tradeoffs of a hybrid-electric vehicle. References: Bailey, G., Mancheri, N. & Van Acker, K. 2017. Sustainability of Permanent Rare Earth Magnet Motors in (H)EV Industry. Journal of Sustainable Metallurgy, 3, 611-626.nBIELLO, D. 2016. Electric Cars Are Not Necessarily Clean. Scientific American. Scientific American, a division of Nature America, Inc.,HICKMAN, L. 2012. Are electric cars bad for the environment. The Guardian. nPUBLISHERS, I. 2008. Hybrid Electric Vehicles Not As Green As They Are Painted (sysy Content Online). Inderscience Publishers nAvailable: www.sciencedaily.com/releases/2008/02/080207094314.htm [Accessed November 27, 2017].

WE261 Developing a National Food Inventory to estimate the Carbon Footprint of the diet Average Spanish. Future requirements and policy recommendations L. Batlle-Bayerl, Universitat Pompeu Fabra UPF / UNESCO Chair in Life Cycle and Climate Change Escola Superior de Comerç Internacional ESCI; A. Bala, UNESCO Chair in Life Cycle and Climate Change (ESCI-UPF) / UNESCO Chair in Life Cycle and Climate Change, Escola Superior de Comerç Internacional ESCI; P. Puluana, Universitat Pompeu Fabra UPF / UNESCO Chair in Life Cycle and Climate Change Escola Superior de Comerç Internacional ESCI; R. Aldaco, Universidad de Cantabria / Department of Chemical and Biomolecular Engineering Dietary patterns have a significant impact on greenhouse gases (GHGs), and dietary choices can increase or reduce the Carbon Footprint (CF) of consumers. Recently, more research has focused on estimating and comparing the CF of different diets; however, high uncertainty is caused by the lack of reliable, available or representative data. The current study discusses this issue on data availability, and it results from our previous study on the CF of the annual food consumption of an average Spanish citizen. To calculate the CF of the average Spanish dietary pattern, a list of food categories with its representative food products was analyzed quantitatively. The aim is to identify bottlenecks for such a quantitative analysis as well as to identify the hotspots from both an environmental and economic point of view. Furthermore, the results will serve as one of the important inputs in laying the foundation for the development of the sustainability boundaries of this study are from cradle inventory (inputs, outputs and emissions) per each food product. The system boundaries of this study are from cradle-to-consumer, and data for all life cycle stages (crop cultivation, farming systems, fisheries, industrial processing, manufacturing, distribution and consumer use) were gathered. Furthermore, food losses and food waste along the whole supply chain were also considered. While total annual emissions, about 1.4 Tn per Spanish citizen, were considered within the uncertainty this research is not an estimate of representative data at a National level. The proxies used to fill the data gaps were considered of good quality. However there is a need to develop inventories for production of certain food products for which there is no inventory data available, as well as other life cycle stages, such as the wholesale & retail and the consumer phase. Furthermore, we suggest the inclusion of environmental data in food policy, for example, adding the CF of food products and dietary patterns within the national dietary guidelines.

Life Cycle Air Emissions External Costs Assessment for comparing Electric and traditional passenger cars P. Girardi, P.C. Brambilla, RSE Spa / SFE

The scope of this study is to compare the externalities of electric, gasoline and diesel motorisations of an average passenger car (aVW Golf) giving a complementary reading of the results of an LCA. Starting from the results of the NEEDS project, authors present a methodology taking into account: the year of the vehicle; the overall regional externalities; the potential irreversibility, thresholds and risks to essential function. At the global level, the UN Sustainable Development Goals apply to all countries and provide a foundation for such a governance framework. We are all consumers and beneficiaries of natural capital, but it seems clear that SETAC members can also inform the debate. As environmental scientists we can contribute to developing an understanding of criticality – the tipping points for ecosystems, in particular where these relate to chemical pressures. There are opportunities to work with economists to ensure natural capital accounting and valuation is accurate and measurable, preferably quantitative. One of the strongest themes was that we all need to improve our story telling across multiple disciplines and institutions.

WE262 Life Cycle Costing: methodological description and application B. De Caevel, J. Dulbecco, RDC Environment; A. Ciroth, GreenDelta; T. Huppertz, I. Descos, RDC Environment; J. Garcia, SCORO LCA

The complexity of production processes and products combined with an increasing demand for environmental knowledge has created the need to monitor and analyze not only the production phase, but also all upstream and downstream costs. Thus, producers are seeking to minimize the overall cost of their product generated throughout the life cycle. The private consumer, on the other hand, seeks to compare the different investment or purchase options by trying to integrate, in a holistic way, their present costs (acquisition prices and associated taxes) and future costs (use, maintenance and end of life costs) of the product with which they want to end up. Life Cycle Costing (LCC) is the typical tool to meet these objectives as it allows to analyze the cost structure of a specific object throughout its life cycle. Due to their convergent approach over life cycle, there are some complementarities between LCC and Life Cycle Assessment (LCA). A combined application of the two approaches could help economic actors and decision makers to find cost-effective solutions, while minimizing their negative external impacts. The external costs evaluation for the analysed vehicles shows that Electric Golf performs better in terms of external costs, mainly thanks to the minor costs due to Climate Change. As regard regional externalities, the external costs due to emissions in Italy make the electric vehicle even more competitive than considering the overall regional externalities.

WE263 Life Cycle Costing: methodological description and application B. De Caevel, J. Dulbecco, RDC Environment; A. Ciroth, GreenDelta; T. Huppertz, I. Descos, RDC Environment; J. Garcia, SCORO LCA

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WE264 Pizzazz: it is dangerously delicious! K. Styllianou, University of Michigan - School of Public Health / Environmental Health Sciences; V. Nguyen, University of Michigan / Department of Computational Medicine and Bioinformatics; V.L. Fulgoni III, Nutrition Impact, LLC; O. Jolliet, University of Michigan

The saying, we are what we eat, is true! Diets are key for human health, more than 10 million deaths/year worldwide are attributable to dietary risk factors. A challenge food Life Cycle Assessment (LCA) faces is that nutrition, a dominant factor, cannot be considered. This leads to results which are not comparable with other LCA studies. To overcome this limitation, we combined LCA with a large-scale consumer survey to evaluate the nutritional impact of the life cycle of a single food item, pizza. This approach allows not only for a comprehensive LCA but also to estimate the impact on critical dietary risks.
primarily focuses on single ingredient items while a large portion of modern diets is comprised of mixed dishes, a mixture of ingredients, which often remains unexplored. We propose a framework for evaluating mixed dishes in LCA that considers nutritional health impacts and demonstrates its application on pizza. We develop 14 marginal nutritional characterization factors (CFs) that cover major food groups and nutrients and allow the assessment of nutritional health effects in LCA. CFs are estimated by coupling age- and gender-adjusted nutritional-specific incidence rates with risk ratios and severity factors from the Global Burden of Disease, measuring benefits (+) and impacts (-) in avoided μDALY/g. To evaluate the environmental impacts, we deconstruct pizza into “basic ingredients” using the USDA Standard Reference 28 database with a resolution of 3,200 single- and multi-ingredients that we further deconstruct. Ingredients are then linked to life cycle inventory (LCI) datasets from the Ecoinvent v3.2, the World Food LCA Database v3.1, and the ESU World food database. We evaluate impacts using Impact World+. Nutritional CFs for food group and nutrient range between -8 (sodium) and 57 (omega-3 from seafood) avoided μDALY/g. Human health scores for pizzas range from -35 avoided μDALY/serving pizza with extra meat to 2 avoided μDALY/serving pizza with no cheese. For the environmental impact assessment, global warming estimates vary from 0.06 (pizza with no cheese) to 0.20 (pizza with extra meat) kg CO2 eq/serving, corresponding to -0.04 and -0.17 avoided μDALY/serving, respectively. When it comes to pizza, environmental emissions further enhance nutritional health impacts. Nutrition can dominate the human health and should be considered in food LCA. We have developed an approach bringing together environmental and nutritional health effects in a comprehensive way that to provide clear and precise FU is to compare protein sources based on the nutrient content of feed. Protein requirements and recommendations enable good practice. The requirements and guidelines for protein intake are linked to human health and should be considered in food LCA. We have developed an approach bringing together environmental and nutritional health effects in a comprehensive way that helps to understand the health impacts of diets.

WE265 The impact of supplemented amino acids in animal feed - a new Life Cycle Assessment approach using the Protein Quality Index as functional unit for comparing protein sources

A. Wojciechowski, Evonik Technology & Infrastructure; M. Binder, Evonik Nutrition & Care GmbH

Proteins, which are made up of amino acids (AA), are essential for human health. Most of AA can be synthesized by the body and 8 of them are called "Essential Amino Acids" (EAA) because they cannot be produced by human or animal and it is crucial to get them through food or feed intake. AA are also used as supplements in animal feed, providing the option to reduce the protein content of feed. Protein production has a major impact on the environment: it is responsible for ~14.5% of all human health impacts, global warming estimates vary from 0.06 (pizza with no cheese) to 0.20 (pizza with extra meat) kg CO2 eq/serving, corresponding to -0.04 and -0.17 avoided μDALY/serving, respectively. When it comes to pizza, environmental emissions further enhance nutritional health impacts. Nutrition can dominate the human health and should be considered in food LCA. We have developed an approach bringing together environmental and nutritional health effects in a comprehensive way that helps to understand the health impacts of diets.
framework which links measurable metrics of impact to value terms that resonate with the public, and reflect value statements made by the community. When the trade-offs are considered, it is important to consider the needs, demographics and vulnerabilities of a diverse population. Sustainability and ES concepts can and should be support environmental decision making; the application of threshold criteria ensures ‘strong’ sustainability in which environmental considerations are not considered in isolation or as a means to achieve other ends. Refinement of the framework which guides stakeholders to consider the extent to which they prioritize impacts to all (rather than just a narrow sub-set) of their values provides for a balanced public comment process, less subject to single- or narrow-issue lobbying. Identification of the risks and benefits of most interest to stakeholders also can support negotiation and optimization of alternatives under consideration, support collaborative design of more sustainable options and help inform the design of a long-term monitoring plan that addresses community values. The goal should be to envision a sustainable approach from the beginning of a project with collaborative input from a large group of stakeholders, supporting informed, transparent, and balanced decision making that protects services of importance to the community. Tools and approaches, and the path forward, will be discussed.

Salt of the earth - causes, consequences and management of salinization of surface freshwaters, groundwaters and soils (P)

WE269

Effects of long-term exposure to increased salinity in the amphibian skin bacterium Erwinia toletana

A.C. Gabriel, University of Aveiro / Biology; I. Lopes, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro; S. Costa, Universidade de Aveiro / Department of Biology & CESAM, 3810-193 Aveiro; I. Henriques, Universidade de Aveiro / Departamento de Biologia CESAM

Amphibians constitute the class of vertebrates with the highest proportion of endangered species: chemical contamination being a main factor for its populations and species decline. The skin bacterial community of amphibians may help them to cope with such chemical contamination. If skin bacteria may increase its tolerance to chemicals, then they could be used for bioaugmentation in amphibians to help them cope with contamination. This work aimed at assess if an amphibian skin bacterium could increase its tolerance to NaCl after long-term exposure to low levels of salinity. Erwinia toletana, isolated from the skin of Pelophylax perezi, was selected as the model species. Clonal populations of this bacterium (5 replicates) were exposed for 46 days to LB medium (Et-LB) or to the effective concentration of NaCl causing 10% of growth inhibition (Et-NaCl; 18 g/L). To assess the capacity of recovery from long-term exposure to NaCl, after the 46 d period, Et-NaCl was transferred to LB medium and cultured for a period of 16 d (Et-R). The isolate also continued to be cultured in LB medium for further 16 d. The tolerance of ancestral and evolved populations to NaCl was assessed by exposing them to 6 NaCl concentrations (5, 10, 15, 20, 25 and 35 g/L) plus a control (LB medium).

Effects of NaCl on bacteria growth and metabolic mechanisms (as degradation of carbon compounds) was monitored. Genotypic alterations were assessed using a PCR-based molecular typing method (BOX-PCR). Results of growth shown that long-term exposure to NaCl slightly increased the tolerance of E. toletana to this salt, EtEC₅₀ for growth were: 22.5 g/L (8.64-36.4) for Et-LB; 30.3 g/L (23.7-37.4) for Et-NaCl, and 26.1 g/L (19.3-32.9) for Et-R. Though, as confidence limits overlapped, tolerance increase was not considered significant. Furthermore, differences in metabolic processes were observed between Et-LB and Et-NaCl, suggesting the use of different carbon sources. This could be associated with the activation of detoxification mechanisms or energetic demanding mechanisms to cope with osmotic stress. Genotypic alterations were not observed, indicating that E. toletana increased tolerance to NaCl could be due to membrane plasticity mechanisms to cope with osmotic stress. The tendency shown by E. toletana to acquire increased tolerance to low levels of salinity could constitute a promising bioaugmentation tool in amphibian’s skin, aiming the improvement of these organisms tolerance to chemicals.

WE270

Impacts of agriculture brackish effluents on growth and metal accumulation parameters of Pelophylax perezi (P)

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Salinity is considered one of the most important factors of soil and water degradation worldwide. Anthropogenic salinization due to the use of low quality water and over-fertilization leads the existence of saline effluents that degrade water quality and constrain plant growth and crop production. However, in saline wetlands salinity is not undesirable, but a proxy parameter to maintain the singularity of the ecosystem. In these environments the existence of a diversity of habitats is closely related to gradients of soil salinity and moisture, with extremely saline sites, brackish sites, wetter sites and drier sites. While agricultural saline effluents may salinize normal soils, they can degrade saline wetlands by decreasing the native soil salinity. This work reports changes in ecosystem structure and diversity in a saline wetland adjacent to the Mar Menor saline lagoon (SE Spain). Species cover, soil salinity, and the groundwater level were monitored in two 2-years periods with a difference of 13 years between them. The results indicated an elevation of the water table throughout the 13-year period, which was attributable to brackish water flowing from areas with intensive agriculture. The latter led to an increase of flooding periods, a decrease of soil salinity in the near the saline sites and an increased in the least saline ones. Following these changes, damages in protected habitats were observed, due to the proliferation and increase of biomass of several species. Sarcocornia fruticosa, Phragmites australis and Juncus maritimus strongly expanded at the wettest sites, which led to the disappearance of the original zonation pattern and the homogenization of the ecosystem. Bare areas, necessary for nesting and feeding of several bird species, disappeared. According to the results obtained, a decrease of soil and water salinity was one of the main factors contributing to the degradation of the saline wetland leading to an exacerbated growth of some species and a decrease of habitat diversity. In this case, the low salinity of the effluents reaching the wetland was a problem, not an advantage.

WE271

Context dependent toxicity - do ecological interactions alter the effects of salinity on stream macroinvertebrate communities?

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The effect chemicals on populations and communities have long been noted to vary between different studies although the mechanism(s) for this variation is unclear. Research has examined variability associated with chemical and physical environments (e.g. bioavailability, co-occurring contaminants) and ecotoxicological and physiological (e.g. temporal and spatial variability in species’ sensitivity). Less consideration has been given to ecological mechanisms including those mediated via indirect effects such as competition and predation. For example, a given population of a species may be able to persist in the presence of a particular level of contamination, but this persistence may be dependent on competition and predator prey interactions, and the relative fitness of these taxa at that level of contamination. Here we report the results of a mesocosm experiment that examined the effects of biotic interactions on salinity effects. We examined effects across a broad salinity gradient using ‘sensitive’ communities collected from a low salinity site (~80 μS/cm) and ‘tolerant’ communities (collected from a high salinity site ~1600 μS/cm). This was examined using a mesocosm experiment consisting of 32 independent re-circulating 1000 L mesocosms. Controls (100 μS/cm) and salinity treatments (500, 1000, 2500 and 5000 μS/cm) these were replicated 4 fold and were crossed in an orthogonal design with the source biota (stream macroinvertebrates and microbes) either from: (1) a low salinity site only or (2) both low and high salinity sites. The experiment is based on the logic that if salinity increases at a site, organisms have the potential to migrate from higher salinity sites within the same region. Thus the organisms from the (previously) low salinity site would have to be able to tolerate both the increase in salinity and ecological interactions with organisms from higher salinity sites. We observed differing effects of salinity on the macroinvertebrate community from the low salinity site depending whether these biota were co-inhabiting with biota from a high salinity site. Such context dependent toxicity deserves greater consideration in studies of the effects of chemicals on populations and communities.

WE272

Challenges in developing a water quality guideline for water hardness

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Increases in salinity, or the total ionic content, of water can be toxic to freshwater species, as can shifts in the balance of major ions (Ca²⁺, Mg²⁺, Na⁺, K⁺, HCO₃⁻, SO₄²⁻, Cl⁻), or increases in the Ca²⁺ and Mg²⁺ content of water alone (i.e. water hardness). Although anthropogenic salinization of freshwater is increasing, virtually no water quality guidelines (WQG) exist for regulating these individual ions or ion mixtures. When there are no WQGs for ions, their use is based on potentially toxic concentrations via effluent, produced waters, and saline run-off from various human activities, e.g. coal mining, oil and gas extraction, the use of Ca- and Mg-based road de-icers, and agriculture. Thus, developing a water hardness based WQG for the protection of aquatic life is warranted. Using Canadian protocols for WQG development, we attempted to derive a WQG for water hardness. Relevant studies collected for this project were reviewed for inclusion in the WQG, however, current literature offered several challenges and major data gaps that hindered WQG derivation. Moreover, the background variation of water hardness throughout an exemplary regulatory region of interest also did not support WQG development using traditional methods. These challenges and limitations will be discussed in the context of similar regulations from other jurisdictions, the need to consider additional, practical limitations of regulating water hardness, or major ions in general, recommendations for improved data consistency, and potential regulatory options.
WE273
Prioritization of water quality stressors according to their relative impact on ecological quality of rivers using large-scale field data: salinity first?
E. Berger, Senckenberg Gesellschaft / Department Quantitative Landscape Ecology; R. Schüfer, University Koblenz-Landau; P. Haase, A. Sundermann, Senckenberg
The political aim of achieving good ecological quality of all European water bodies requires knowledge on how to prioritize stressors and human pressures for management based on their relative impact. A challenge thereby is the frequent co-occurrence of multiple stressors. We applied eco-epidemiological approaches to large scale monitoring data from Saxony, Germany, to investigate the relative contribution of different water quality and land-use gradients to ecological change. Two approaches were applied: First, water quality gradients (e.g. oxygen, conductivity, phosphorous and microplottants) and land-use gradients (e.g. % arable and urban catchment land cover, position of wastewater treatment plants) were used as predictor variables in multiple linear regression analysis and hierarchically partitioning with ecological quality indices based on invertebrates (% EPT, MMI, ASPT, BMWP, GSI, SPEAR %) as response variables. Secondly, individual taxon richness was used with respect to different water quality gradients (including also major ions such as potassium, sodium, chloride etc.) were assessed using Threshold Taxa Indicator Analysis (TITAN). The method is based on change point and indicator species analysis and allows the identification of ecological change points that may be used to derive environmental quality criteria. Both regression analysis and TITAN results indicate a high impact of oxygen and salinity, which were allocated as the most severe stressors and salinity (because there are no State standards or USA water quality guidelines). Although observed associations may not be direct causes of ecological impairment, it may be worthwhile to implement legally binding quality standards for these variables. Of the 324 analyzed taxa 23% had change points far below the German orientation value for chloride (200 mg/L) that should not be exceeded to achieve good ecological status according to the water framework directive. Thus, lowering of the concentration value and associated ion concentrations should be considered to protect and restore stream biodiversity. Moreover, the results suggest that preventing release of poorly treated wastewater should be prioritized over up-grading of well-functioning treatment plants.

WE274
Estimating protective potassium concentrations for freshwater mussels, a taxon of global conservation concern
T. Augspurger, U.S. Fish and Wildlife Service / Ecological Services
Globally, there are about 620 species of freshwater mussels (Family Unionidae), and IUCN lists 28 species as extant and 106 as endangered or critically endangered. Mussels are among the most sensitive freshwater water organisms to toxicity from chloride and potassium, and the environmental relevance of these is increasing with sea level rise and brine discharges. Average potassium concentrations in relatively unpolluted streams of North Carolina (USA) range from 0.2 to 2 mg/L. An industrial effluent with potassium averaging 504 mg/L and proposed for discharge to a stream with endangered mussels necessitated derivation of protective potassium limits (because there are no turbid water standards in USA water quality criteria for potassium). From the literature, we compiled potassium 96-h EC50s (with endpoints of lethality or immobilization) for mussels and retained those with > 90% control survival, measured test chemical concentrations, and acceptable test water quality. Five EC50s ranged from 31 to 48 mg/L at a water hardness of 100 mg/L as CaCO3, and we applied the North Carolina guidance of one potential protective potassium limit because the standard limits are chloride specific. We adjusted the 10 mg/L acute limit to a water hardness of 18 mg/L which is the 5th percentile of the proposed receiving stream (protective most of time because potassium is less toxic as hardness increases). The hardness-adjusted acute water quality guideline of 7 mg/L potassium was recommended as an instantaneous concentration not to be exceeded. Chronic toxicity data for potassium and mussels were available for two studies from 28 to 300 mg/L. Because mussels can live for decades, we used the 300-d test. The geometric mean of the test NOEC (1 mg/L) and LOEC (7 mg/L) yield a chronic value of 2.6 mg/L potassium which was recommended as a monthly average guideline not to be exceeded more than once every three years. We used 32 years of receiving stream flow data to derive a predicted potassium yield a chronic value of 2.6 mg/L potassium which was recommended as an instantaneous protective potassium limit (because there are no State standards or USA water quality guidelines of toxicant and discuss recommendations for quarterly monitoring activities showed that conservation status of the habitat 1150* is improving within SCI FT2350031, but it is still unfavourable in the inner landward areas due to lack of ecological corridor areas, favouring self-regulation processes, between lagoon and mainland. In the past, the project area was occupied by reedbeds in large amounts, now significantly degraded due to increasing of lagoon water salinity, caused by historical human activities (e.g. diversion of rivers with reduction of freshwater supply, inlet and channel excavation). With reduction or disappearance of reedbeds, their contribution to ecosystem services, like supporting numerous biological communities and species, are minimized. The LIFE LAGOON REFRESH project, started on Sept 2017, foresees the restoration of favourable conservation status of habitat 1150* in the northern Venice Lagoon and the recreation of favourable habitats for faunal species of community interest. The project actions involve: diversion of a freshwater flow from the Sile river into the lagoon (necessary for the recreation of the typical salt gradient of buffer areas between the lagoon and mainland); restoration of intertidal morphology through the implementation of structures properly arranged to slow down the freshwater dispersion and to favour reed development; planting of Phragmites australis to accelerate the development of the reedbeds; transplantation of small dumps of seagrass species of the habitat 1150*, suitable to accelerate the recolonization by aquatic plants of low-salinity environments. The project aims to exploit the statistically diluted effluents and synthetic solutions (e.g. NaCl-CaCl2) of similar salinity. These variables were greater from industrial wastewater than the comparable brine solution. The tolerance of Festuca rubra L. (fescue) and Trifolium pratense L. (clover) were investigated for 8 weeks under hydromorphic conditions to compare their growth in brine effluents from tertiary recovery operations. Experiments further compared serially-diluted effluents and synthetic solutions (e.g. NaCl-CaCl2) of similar salinities. There were different growth responses to the wastewater and saline solutions among both plant species. F. rubra was exhibited a significant higher survival percentage than T. pratense. After four weeks of exposure, T. pratense exhibited higher sensitivity and lethality. Interestingly, biomasses of both plants were greater from industrial wastewater than the comparable brine solution. The restoration of salinity gradients will also contribute to increase biodiversity within the Natura 2000 network site, and to help achieving targets defined by the 2020 Biodiversity Strategy.

WE276
Comparing the growth of fescue and clover plants in petroleum industrial effluents and solutions of similar salinity
P. Srihakhunsuk, University of Strathclyde / Department of Civil & Environmental Engineering; C. Knapp, J. Renshaw, University of Strathclyde / Civil and Environmental Engineering
Effluents (produced and flow-back waters) from the petroleum industry have been between 500 and 1000 mg/L salt and toxicity to the environment, particularly related to chemical composition and salinity. The purpose of this study was to investigate whether their toxicity is any greater than exposure to solutions of similar salinity. The tolerance of Festuca rubra L. (fescue) and Trifolium pratense L. (clover) were investigated for 5 weeks under hydromorphic conditions to compare their growth in brine effluents from tertiary recovery operations. Experiments further compared serially-diluted effluents and synthetic solutions (e.g. NaCl-CaCl2) of similar salinities. There were different growth responses to the wastewater and saline solutions among both plant species. F. rubra was exhibited a significant higher survival percentage than T. pratense. After four weeks of exposure, T. pratense exhibited higher sensitivity and lethality. Interestingly, biomasses of both plants were greater from industrial wastewater than the comparable brine solution. The restoration of salinity gradients will also contribute to increase biodiversity within the Natura 2000 network site, and to help achieving targets defined by the 2020 Biodiversity Strategy.

WE277
Contribution to the salinization risk assessment, under drought conditions, in the Alqueva irrigation area (South Portugal)
In the northern Venice Lagoon (SCI FT2350031) holds several Annex I-listed habitats of the Habitats Directive, such as the habitat type 1150* (Coastal lagoons). Recent monitoring activities showed that conservation status of the habitat 1150* is improving within SCI FT2350031, but it is still unfavourable in the inner landward areas due to lack of ecological corridor areas, favouring self-regulation processes, between lagoon and mainland. In the past, the project area was occupied by reedbeds in large amounts, now significantly degraded due to increasing of lagoon water salinity, caused by historical human activities (e.g. diversion of rivers with reduction of freshwater supply, inlet and channel excavation). With reduction or disappearance of reedbeds, their contribution to ecosystem services, like supporting numerous biological communities and species, are minimized. The LIFE LAGOON REFRESH project, started on Sept 2017, foresees the restoration of favourable conservation status of habitat 1150* in the northern Venice Lagoon and the recreation of favourable habitats for faunal species of community interest. The project actions involve: diversion of a freshwater flow from the Sile river into the lagoon (necessary for the recreation of the typical salt gradient of buffer areas between the lagoon and mainland); restoration of intertidal morphology through the implementation of structures properly arranged to slow down the freshwater dispersion and to favour reed development; planting of Phragmites australis to accelerate the development of the reedbeds; transplantation of small dumps of seagrass species of the habitat 1150*, suitable to accelerate the recolonization by aquatic plants of low-salinity environments. The project aims to exploit the statistically diluted effluents and synthetic solutions (e.g. NaCl-CaCl2) of similar salinity. These variables were greater from industrial wastewater than the comparable brine solution. The tolerance of Festuca rubra L. (fescue) and Trifolium pratense L. (clover) were investigated for 8 weeks under hydromorphic conditions to compare their growth in brine effluents from tertiary recovery operations. Experiments further compared serially-diluted effluents and synthetic solutions (e.g. NaCl-CaCl2) of similar salinities. There were different growth responses to the wastewater and saline solutions among both plant species. F. rubra was exhibited a significant higher survival percentage than T. pratense. After four weeks of exposure, T. pratense exhibited higher sensitivity and lethality. Interestingly, biomasses of both plants were greater from industrial wastewater than the comparable brine solution. The restoration of salinity gradients will also contribute to increase biodiversity within the Natura 2000 network site, and to help achieving targets defined by the 2020 Biodiversity Strategy.
major inorganic ions (Na⁺, Ca²⁺, Mg²⁺, K⁺, SO₄²⁻ and Cl⁻), pH and electric conductivity (ECa), was conducted throughout 2017, on water samples collected on four platforms sited in the reservoir. Water quality for irrigation was evaluated considering both the Portuguese regulations and the FAO guidelines. Sodium adsorption ratio (SAR) and soil salinity (ECe) were estimated, in order to assess potential sodium-related soil permeability and crustling problems, as well as, potential yield reductions in the most important crops of the Alqueva perimeter. Higher ion concentration related with water salinization. We selected water samples of atmosphere evaporative demand. Sodium hazard assessment showed slight to moderate risk of reduced infiltration rates, a result that should be taken into account when surface or sprinkler irrigation systems are used. Furthermore, relative yield reductions may be mainly found in horticultural crops, classified as moderately sensitive to sensitive in the salt tolerance scale.

Systems ecotoxicology: application of OMICS data across multiple levels of biological organization in research and risk assessment (P)

WE279 Investigating wildlife diets using high-tech DNA sequencing
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In wildlife risk assessments according to EFSAs (2009), the ingested diet is one of the core factors to define exposure, using default diet compositions in the first tier risk assessment. This risky approach is based on the assumption of the standard refinement parameters which intend to add realism to higher tier risk assessments. Publically available dietary data are often used to refine PD in wildlife risk assessments; however, such data are often variable and/or not representative for the specific risk assessment scenario. Besides such literature data, specifically registration-relevant PD field studies can be conducted. PD values gathered from such data are based on collated factors, study for their inclusion in test toxicity tests. In these studies samples (or parts thereof), are investigated visually by microscope and food items are identified based on comparison with comprehensive reference data libraries and collections of potential diet items. For plant material, the results are mainly presented on a fairly basic taxonomic level and are often related to wildlife risk assessment defined diet fractions which have different default residue levels for all detoxifiable plants or non-detoxifiable plants only. However, this is rather time-consuming and imprecise. Recently, DNA sequencing techniques are increasingly applied for diet composition analyses in ecological science. We initiated an approach using ‘next-generation’ DNA amplicon sequencing to quantitatively assess the diet composition of wild herbivorous mammals, taken from faeces samples collected on arable fields. Data on the relative abundance of each plant species were derived by enrichment and sequencing of a specific DNA region (ITS2 region of the ribosomal DNA) and by comparison to comprehensive plant species DNA databases. The approach has proved to be very useful on identification of relative abundances of plant species from faecal samples. This new genomics approach, its needs and limitations for refined risk assessment will be presented and discussed.

WE280 Design of a Real-Time PCR array to analyze the gene expression in Physella acuta (Gastropoda) in chemical stress and starvation
M. Novo, J. Martinez-Guitarte, UNED / Fisica Matematica y de Fluidos Molecular endpoints are nowadays under study for their inclusion in test toxicity tests. While vertebrate species are usually well-known, there is a lack of information on invertebrates. The study of the latter is complex since their body shape, behavior, and ecology are very diverse, and great differences can be found even within the same animal group. In order to improve our knowledge in putative molecular endpoints and to evaluate some genes as biomarkers, a Real-Time PCR array has been designed for Physella acuta. This species of freshwater snail is used in environmental toxicology studies and it has been proposed as an adequate species for toxicity tests because of its sensitivity to different toxicants and ease of culture. A transcriptome for this species was assembled, by sequencing cDNA libraries from individuals of different developmental stages and exposed to different toxicants. Comparison with database allowed the identification of genes involved in pathophysiology and relevant to toxicant response. We selected 42 of these genes to define their expression under different treatments, in order to assess potential yield reductions in the most important crops of the Alqueva perimeter. Higher ion concentration related with water salinization. We selected water samples of atmosphere evaporative demand. Sodium hazard assessment showed slight to moderate risk of reduced infiltration rates, a result that should be taken into account when surface or sprinkler irrigation systems are used. Furthermore, relative yield reductions may be mainly found in horticultural crops, classified as moderately sensitive to sensitive in the salt tolerance scale.

WE281 Performance of different chemical toxicity assessment tools for aquatic polychaete species
A. Winkler, J. Kalinowski, Center for Biotechnology (CeBiTeC) University of Bielefeld
Chemical toxicity has been assessed in order to assess environmental risk due to their potential to incorporate toxicants, and to be indicators of toxicant exposure. Aquatic polychaetes have been used in environmental risk assessment for about 20 years and are widely accepted as a cost-effective and reliable risk assessment tool. This study compared the performance of three different chemical toxicity assessment tools (Cytotoxicity Test, Acute Bioassay, and Chronic Bioassay) for the assessment of environmental risk to aquatic polychaete species. The study involved the assessment of acute and chronic toxicity of 10 different chemicals (pesticides, heavy metals, and organic compounds) on the polychaete species Arenicola marina. The results showed that the Cytotoxicity Test was the most sensitive tool, followed by the Acute Bioassay and then the Chronic Bioassay. The Cytotoxicity Test was able to detect the toxic effects of all the chemicals at the lowest concentrations, while the Chronic Bioassay was the least sensitive. These results highlight the importance of using multiple assessment tools to accurately assess environmental risk to aquatic polychaete species.

WE282 Environmental risk assessment for soil toxicology studies
M. Roelofs, Vrije Universiteit / Department of Ecological Science; N. van Straalen, Universiteit Utrecht / Ecological and Environmental Biology; K. De Schamphelaere, Ghent University (UGent) / Applied Ecology and Environmental Biology
Over the past decades, the world’s oceans and seas have been influenced by several human induced impacts, including climate change. In the North Sea region, the average sea surface temperature of the water has already risen with 1-2 °C over a time span of twenty-five years and is likely to continue in the future. Understanding the impacts of this changing environmental condition in zooplankton communities is crucial, as alterations in the zooplankton communities can affect entire marine ecosystems. Here, we focus on the potential effects of an increase in temperature on the calanoid copepod species, Temora longicornis, the dominant zooplankton species of the southern part of the North Sea. Since responses to environmental stress are genome-driven, a genetic study on the physiological responses to thermal stress can provide an increased mechanistic understanding and help predict potential responses to climate change in this copepod species. Therefore, we sequenced the whole transcriptome (using RNA-seq technology) in T. longicornis, after being exposed to thermal stress, to investigate gene expression differences as a response to temperature fluctuations. As such, this dataset will provide insights into the mechanisms underlying the observed changes in gene expression. This study is expected to contribute to a better understanding of the physiological responses of T. longicornis to thermal stress, which will be essential for predicting the potential impacts of climate change on marine ecosystems.

WE283 Validating a contamination assessment tool from lab to the field: Folsomia candida exposed to a fungicide-based formulation
T.F. Simoes, S.C. Novais, Polytechnic Institute of Leiria / MARE IPLeiria; T. Natal da Luz, University of Coimbra / Department of Life Sciences, University of Coimbra; J. Renadn, CEFAS / Centre for Environmental and Fisheries Science; J. Römönke, ECT Oekotoxikologie GmbH; D. Roelofs, Vrije Universiteit / Department of Ecological Science; N. van Straalen, Association of Retired Environmental Scientists ARES / Department of Ecological Sciences; M.F. Lemos, Instituto Politécnico de Leiria / MARE IPLeiria
Folsomia candida is a widespread arthropod that occurs in soils throughout the world and has been used as a standard test organism in past decades for estimating the effects of pesticides and environmental pollutants on non-target soil arthropods. This species is among the most sensitive representatives of its taxon, being selected as a genomic model organism for soil toxicity studies. Although laboratory
experiments with a transcriptomics approach are essential to unravel modes of action of chemical compounds, higher-tier studies (e.g., field studies) are crucial as a validation criterion in environmental risk assessment tests, while their ecological relevance is increased when complemented by pertinent information at lower-tier studies (molecular level). Therefore, the main goal of the present study was to validate the mode of action of a commercial fungicide formulation in *F. candida* under a number of field and laboratory scenarios, by targeting specific molecular biomarkers retrieved from a previous laboratory and field study on the previous data of survival and reproduction effects in *F. candida* exposed to a commercial formulation of the fungicide chlorothalonil (40% Bravo®500) in a natural agricultural soil under laboratorial conditions, organisms were now exposed under field conditions for 4 days to the same concentration as for laboratorial exposure (causing a 75% reductive on reproduction) and the Predicted Environmental Concentration (5 mg a.i./kg). Invertebrates were previously cultured in laboratory and simultaneously 12 replicate soil cores per treatment (including control) were collected from the field and deaerated. The cores were placed back in the field and 220 organisms (10-12 days old) were added per replicate core. Field contamination was made by spraying after a 3 hours acculation period to the field by the organisms. RNA was extracted from each pool of organisms using the TRIZOL® methodology. According to previous laboratorial “omics” results with the same assay, a set of specific genes were selected for a targeted gene expression analysis by qRT-PCR, corresponding to key genes of affected biological pathways including glutathione metabolism, oxidation-reduction, body morphogenesis and reproduction. This work contributes with a set of molecular biomarkers which can be used to develop a more effective set of tools to assess the early effects of each fungicide formulations in a real scenario of soil contamination.

**WE284**

Proteome response of Chironomus riparius under exposure to the neurotoxic insecticides Spinosad and Indoxacarb

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The development of quantitative methodologies in proteomics opened new doors for their potential applications within environmental sciences. Since proteins are the functional units of cells, the proteome of an organism at a given time and at a given condition reflects its current state. In this sense, any protein profile changes in response to toxicants may reveal their molecular targets and/or specific stress-response mechanisms, and thus can be used as potential early warning biomarkers of toxic exposure. In this study, the potential of proteome changes as an early warning indicator for pesticide exposure in *Chironomus riparius* (Meigen) was evaluated using as model compounds two neurotoxic pesticides with distinct modes of action, spinoasod and indoxacarb. *Chironomus riparius* third-instar larvae were exposed to three concentrations of each pesticide and iTRAQ methodology was performed to relatively quantify protein expression changes between exposed and non-exposed organisms. For each exposure period, the pesticides expression triggered different responses at the proteome level. Changes caused by spinoasod were more noticeable than for indoxacarb exposure. Our results revealed a general decrease in the expression of globin proteins with the increase of spinoasod concentration. Additionally, for spinoasod, a significant decrease in the expression of an actin and a globin protein were also observed. Additionally, for spinosad, a significant decrease in the expression of an actin and a globin proteins with the increase of spinosad concentration. The observed decrease of actin and globin proteins might reflect toxic stress-related responses, such as reduced metabolic rates, as a result of the exposure. Further research is needed to validate these findings in different field conditions, in order to determine the potential of proteomics as a tool for early warning of environmental stress.

**WE288**

Assessing Cu impacts on freshwater diatoms: biochemical and metabolic responses of *Tabellaria flocculosa* (Roth) Kützing

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Metals are a recognised threat to aquatic organisms but the impact of metals such as copper (Cu) on benthic freshwater diatoms is poorly understood, even if diatoms are commonly used as water quality indicators. Our study aimed to elucidate the cellular responses of diatoms to Cu toxicity. The freshwater diatom *Tabellaria flocculosa* (TFLQ), isolated from a Cu contaminated stream, was exposed to 0.3, 6 and 10 µg Cu/L, and the tolerance level and the cellular targets were studied using biochemical, physiological and metabolic approaches. Cu was already toxic to *T. flocculosa* at concentrations common in environments which are usually not considered to be contaminated (0.3 µg Cu/L), and toxicity increased with Cu concentration. Strategies to cope with Cu varied with the level of Cu stress. Under Cu impact, the metabolome of *T. flocculosa* changed significantly, especially at high concentrations (6 and 10 µg Cu/L). Cu toxicity was counteracted by increasing extracellular immobilization (EPS, frustules), antioxidant (SOD, CAT) and detoxifying (GSTs) enzymes activity and low molecular weight antioxidants (GSH). These mechanisms were up-regulated by a higher energy production (ETS activity, use of sugars and lipids). At the highest Cu concentration (10 µg/L), these metabolic processes were specially enhanced in an attempt to restrain the oxidative stress generated by high intracellular Cu concentrations. However, these mechanisms were not able to fully protect cells, and damage in membranes and proteins occurred. Moreover, the decrease of hydroxylamine and unsaturated FA and the increase of saturated FA, 2-palmitoylglycerol, glycerol and dimers compounds should be tested as new specific markers of Cu toxicity in future studies. This information can support the prediction of diatom behaviour in different Cu contamination levels, including highly impacted environments, such as mining scenarios, and may assist in environmental risk assessment policies.

https://www.dropbox.com/s/8b1jsezdiqx3rmw/graph%20abs%2020?dl=0

**WE287**

Non-targeted approach to identify metabolic perturbations in gilt-head bream liver and brain exposed to benzophenone-3

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Benzophenone-3 (BP-3) is a widely used organic UV filter to protect humans and materials from damage by UV irradiation. The extensive use of BP-3 has led to its ubiquitous occurrence in the aquatic environment, causing an ecotoxicological risk to organisms. Although some studies reported adverse effects on both invertebrates and fishes, further research needs to be done in order to assess its molecular and physiological effects, and modes of action. Therefore, in the present work, we investigated metabolic perturbations in juvenile gilt-head bream (*Sparus aurata*) exposed over 14 days via the water to BP-3 (50 mg/L). Liver and brain were collected prior to dosing and on exposure days 2, 4, 7 and 14 from control (n=10) and exposed (n=10) animals. Samples were flash frozen and stored at -80°C until analysis. Methanol:chloroform (80:20, v/v) mixture was used for non-selective extraction of fish tissues and subsequent non-target analysis was performed by means of UHPLC-Oriibtrap MS in positive and negative modes with both C18 and HILIC separation. Metabolites were identified using Compound Discoverer (Thermo) interfaced to M/Zmine and the statistical data treatment was carried out with R software. Mortality was not observed during the experiment, and no statistical changes in fish weight, fish length, condition factor and hepatic somatic index were observed regardless of tank or dosing period. First of all, using sequence quality control samples, data had to be corrected to remove the effect of injection order. On the other hand, since some metabolites were significantly affected by exposure, a targeted approach to identify metabolic perturbations in gilt-head bream liver and brain exposed to benzophenone-3 gilt-head bream, non-target metabolomics -Acknowledgements- This work was financially supported by the Ministry of Economy and Competitiveness through the project CTM2014-56628-C3-1-R. H. Ziarrusta is grateful to the Spanish Ministry and L. Mijangos to the Basque Government for their predoctoral fellowships.

**WE268**

METABOLIC ALTERATIONS IN LIVER AND GONADS OF FISH

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Effluents from pulp and paper mills are internationally recognized as containing toxic substances with properties to alter the reproductive capacity of fish. The objective of this study was to evaluate the metabolic modifications (using metabolomics tools) of fish gonads and liver (Danio rerio) exposed to effluents from two different paper industries. The embryonic development of embryo of respective fish. The effluent was diluted by a factor of 1/25. The fish (10 males and females) were exposed to the effluent, and males and females were kept separated for 7 days at 28°C and a light / dark cycle of 12/12 in 3.5 L tanks. After the pre-exposure period, males and females were mated (1 couple per Becker containing a net at the bottom) and monitored for 6 days with daily water/pollutant renewal and were fed twice a day. The eggs were collected and placed on plates containing the test agent, kept protected from light. The fish were sacrificed for liver and gonads removal, from which a homogenate was prepared. Then, extraction was performed with chloroform/ methanol/water (3: 2: 1). The extract
obtained was prepared for gas chromatography. For the exposed fish, there was a significant reduction in egg production, 65%. A high percentage of the eggs presented dark staining, which are not viable. Chromatography analysis revealed significant changes in the amino acid, sterol and fatty acid profile in both tissues, liver and gonads. The results showed a strong impact on the metabolism, egg production and embryo development for the studied fish, which point to the alteration of their reproductive capacity.

WE289 Developing biomarkers of sewage effluent exposure in the freshwater amphipod Gammarus fossarum

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Pesticides, pharmaceuticals, industrial chemicals and complexing agents coming from sewage effluents, can be detected in the aquatic environment in very low concentrations. They are able to impact ecological communities, causing biological alterations in many species. Although biomarkers in vertebrates have long been applied, attempts to monitor dysfunctions in invertebrates using orthologous genes have produced inconsistent results. Gammarids are a group of amphipods that have been shown as very sensitive to pollution, having been used in various studies for toxicity evaluation of river waters through exposure to many different chemicals. However, to date most studies have been focused on specific life-cycle stages, potentially missing complex interactions among expressed genes not involved in development. The aim of this study is to provide a set of new transcriptomic and metabolomic markers in Gammarus fossarum. After validations in further studies, the new biomarkers found in this project could be used to evaluate the state of aquatic ecological niches and the efficiency of wastewater treatment plants (WWTPs). Amphipods were sampled from a freshwater stream located in Elgg, Switzerland in September 2017, using standard kick-net method. This stream flows through an industrial WWTP, steadily exposing the whole fauna and flora to a range of pollutants. Sampling was performed 50 m downstream of the WWTP and 50 m upstream, as reference site. Five biological replicates and five technical replicates for both males and females, sampled upstream and downstream were used to get 20 samples containing total RNA. A subsample of amphipods was collected for metabolomic analysis and additional samples were fixed to record population markers, such as sex ratio, fecundity rate and potential interspecies phenotypes. The RNA samples have been sequenced by Illumina Genome Analyzer. A differential expression analysis will be conducted to identify significantly different genes between upstream and downstream populations. Subsequently, a comparison between transcriptomic and metabolomic markers will be carried out, allowing a better understanding of the biological functions impaired after amphipods wastewater exposure. A biological pathway analysis will be also performed on the differentially expressed genes, allowing a correlation of the impaired molecular pathways after chronic exposure to water pollutants with the chemical mixture found in the wastewater effluent.

WE290 Optimising the algal toxicity test towards generation of multi-omics data and adverse outcome pathway discovery

S. Schade, Birmingham University / Biosciences; N. Taylor, Cambridge Environmental Assessments (CEA) / Ecotoxicology; J. Zhou, S. He, University of Birmingham / School of Chemistry; E. Butler, Unilever; G. Hodges, Unilever / Safety and Environmental Assurance Centre SEAC; J. Colbourne, M.R. Viant, University of Birmingham / School of Biosciences

The adverse outcome pathway (AOP) concept represents a framework to organize mechanistic understanding of toxicological interactions by causally linking critical molecular key events (KE) to apical endpoints relevant for chemical risk assessment. Currently, only few methodologies can be considered for an accurate and reliable discovery and quantification of KEs in an exhaustive approach, commonly requiring sustained research effort. In this context, the objective of our presented proof-of-concept study was to showcase the identification and characterisation of molecular KEs from the molecular stress response of Chlamydomonas reinhardtii to toxic insult, applying a multi-omics approach to provide a high-throughput genome-wide scaling multi-omics technology. The approach towards achieving this end was a suite of untreated (direct-infection mass-spec, DIMS; RNA sequencing) and targeted (LC-MS/MS, -UV, qRT-PCR) metabolomics, lipidomics and transcriptomics technologies. This methodology enabled us to profile the concentration- and time-response profiles of molecular signatures from algae exposed to non-specific mechanism (baseline toxicity) and target-specific mechanism (carotenoid biosynthesis inhibition) toxicants. To enable this work, a rigorously controlled algal culturing and testing system was optimised regarding growth rate, final cell density, pH stability, cell cycle synchronisation, reproducible exposure to volatile chemicals, and rapid quenching and harvesting of biomass for omics data collection. Furthermore, a unique multi-phase experimental design was developed for rapid identification (untargeted), characterisation and verification (targeted) of putative KEs over a time-course design. Multi-omics data from toxicant-exposed C. reinhardtii were collected and initial progress made towards computational analysis, putative KE designation, and targeted verification of identified biomarkers. With this study, a powerful experimental approach for hypothesis-free KE discovery and AOP hypothesis is being developed, employing omics-driven algal phenotyping to advance the integration of omics data into AOP development and ultimately, to provide mechanism-based support for regulatory decision-making in environmental risk assessment.

WE291 Evaluating interactive toxic effects of copper and lead on marine mussels: molecular to physiological consequences

C. Crowther, V. Sharma, Plymouth University; A. Turner, Plymouth University / Food Safety; A.N. Jha, Plymouth University / Biological Sciences

It is increasingly recognised that anthropogenic contaminants are not isolated in their threats to the aquatic environment. Recently there has been a shift towards measuring the effects of exposure to low-concentrations chemical mixtures under chronic conditions to predict outcomes on the ecosystem. Adopting an integrated approach the aim of this study was to assess the interactive effects of copper (Cu) and lead (Pb) either alone or as a mixture at various levels of biological organisation, ranging from molecular to individual levels. The combination of proteomics, molecular and physiological measures with bioinformatics adopted in this study will allow a model of mixture exposures to be created which can be translated to early warning indicators within the marine environment. Mytilus galloprovincialis were exposed to a range of concentrations of Cu (5, 32 μg/L) and Pb (5, 25 μg/L) both individually and in a binary mixture. After a 14 day exposure, a number of physiological and molecular parameters were assessed. This included: measurements of 'clearance rate', activity of metallothionein and activity of specific thioredoxin PRX1, and a lipid peroxidation assay was performed. This hypothesised AOP allowed development of targeted assay data, for the selected biomarkers of copper and lead showed an increase in acetylcholinesterase activity, this treatment was significantly higher than both the highest copper binary mixtures. The analysis of protein profile is in process which should provide the potential functional effects of exposures of these two environmentally relevant metals, either alone or in combination. Furthermore, once this study has determined the chemical interactions between binary metals and the mussel’s proteome this could pave the way for further omics being performed and adapted for the ability to create early warning environmental indicators, not only for the environmental health but also for human health.

WE292 The Identification of Toxicological Markers in Adverse Outcome Pathway Discovery in Chlamydomonas reinhardtii

G. Reynolds, Unilever / Safety and Environmental Assurance Centre SEAC; S. Schade, Birmingham University / Biosciences; N. Taylor, Cambridge Environmental Assessments (CEA) / Ecotoxicology; J. Colbourne, M.R. Viant, University of Birmingham / School of Biosciences

Current regulatory toxicity testing methods have become unsuitable for the thorough assessment of chemicals for commercial use, as lack of insight into toxicological mechanism prevents accurate predictive risk assessment. Adverse outcome pathway (AOP) development to address these weaknesses has become a major focus for regulatory toxicologists. AOPs provide a framework for collating mechanistic data from a diverse range of methodologies, including in silico and in vitro approaches, for use in regulatory decision making. The aim of this work was to develop and test a more comprehensive experimental design, for the targeted characterisation of key events in the toxicological response of Chlamydomonas reinhardtii upon herbicide exposure, thereby contributing to the development of a quantitative AOP. Here we present how an initial hypothesis for an AOP was created from available laboratory evidence, with focus on ‘omics and multiple-endpoint assay data, for the selected herbicide, norflurazon. This hypothesised AOP allowed development of targeted assays for investigation of predicted key events in a time- and concentration-response methodology. LC-UV was used to monitor suspected toxicological markers of the carotenoid biosynthesis pathway (phytoene, phytofluene, b-carotene). qPCR was used to identify differential mRNA expression of chloroplast-specific thioredoxin PRX1, and a lipid peroxidation assay was applied for determining downstream effects of non-specific oxidative stress. A concentration- and time-dependent response in phytene accumulation was observed, whilst concentration dependent b-carotene depletion was shown at later times. Both assays were released for the tested dose within an hour of exposure, whilst lipid peroxidation occurred between 4 and 24 hours post-exposure. Significant ($p < 0.01$) effects on cell number, an adverse outcome, were observed at 2000μg/L after 24 hours. This study highlighted the necessity for use of synchronous algal cultures for approach the aim of this study was to assess the interactive effects of copper (Cu) and lead (Pb) either alone or as a mixture at various levels of biological organisation, ranging from molecular to individual levels. The combination of proteomics, molecular and physiological measures with bioinformatics adopted in this study will allow a model of mixture exposures to be created which can be translated to early warning indicators within the marine environment. Mytilus galloprovincialis were exposed to a range of concentrations of Cu (5, 32 μg/L) and Pb (5, 25 μg/L) both individually and in a binary mixture. After a 14 day exposure, a number of physiological and molecular parameters were assessed. This included: measurements of 'clearance rate', activity of metallothionein and activity of specific thioredoxin PRX1, and a lipid peroxidation assay was performed. This hypothesised AOP allowed development of targeted assay data, for the selected biomarkers of copper and lead showed an increase in acetylcholinesterase activity, this treatment was significantly higher than both the highest copper binary mixtures. The analysis of protein profile is in process which should provide the potential functional effects of exposures of these two environmentally relevant metals, either alone or in combination. Furthermore, once this study has determined the chemical interactions between binary metals and the mussel’s proteome this could pave the way for further omics being performed and adapted for the ability to create early warning environmental indicators, not only for the environmental health but also for human health.
**WE293**

Effects of water-borne benz[a]pyrene on early-life stages of the fathead minnow (Pimephales promelas)

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Polyphenolic aromatic hydrocarbons (PAHs) are a class of ubiquitously distributed environmental pollutants that mainly originate from petrogenic and pyrogenic sources such as combustion of fossil fuels and other organic material. Various PAHs, including benz[a]pyrene (BaP), have been demonstrated to cause a wide range of effects in exposed wildlife, including alterations of immune responses, impaired development and reproduction, as well as mutagenesis and carcinogenesis. Most studies to date, however, have used comparably high exposure concentrations, dietary routes of exposure or intrauterine injection to administer BaP, and knowledge of low-dose effects at concentrations around water solubility (approx. 4 µg/L) is generally limited. This route of exposure, however, must be considered highly relevant in light of the distribution of PAHs even into remote aquatic systems. To bridge this knowledge gap, early-life stages of the fathead minnow (Pimephales promelas) will be exposed to waterborne BaP as a model compound to characterize toxicity pathways that drive the sensitivity of early-life stage-fish to PAHs. Molecular responses at the whole transcriptome, proteome and metabolome level will be investigated at the swim-up stage, and quantitatively correlated with effects on apical growth, survival, development, histopathological, and biochemical endpoints 28 d post-hatch. The data generated within this experiment will help to better understand the relevance of aqueous exposure to BaP specifically, and PAHs in general, and provide important insights into the relevance of molecular responses in early-life stages as early-warning biomarkers for apical outcomes in juvenile and/or adult fish. This study is part of the EcoToxChip project (@ecotoxchip).

**WE294**

SETAC OMICS Interest Group

B. Campos, Unilever R&D / Environmental Chemistry

**Epigenetic and evolutionary toxicology: from mechanisms to risk assessment (P)**

**WE296**

Epigenetic effects in Daphnia magna by characterizing quantified abundance of global methylation, gene expression and histone modifications

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Daphnia magna is used in toxicology and environmental science as a monitor for ecosystem health. Epigenetic analysis is enabled by the genome of the closely related D. pulex. Epigenetic mechanisms allow gene regulation in a developmentally dynamic and environmentally stochastic environment. The best studied epigenetic mechanisms are methylation forms on cytosines in a CpG context and post-translational modifications (PTMs) on histone proteins attached to DNA. The global abundance or change of 5-methyl-cytosine (5mC) may indicate epigenetic reactions to environmental stimuli, since these methylation forms facilitate in the regulation of gene expression the change in expression can be detected and compared. The developed PyzHMM code give insight on how these proteins modifications regulate gene expression and crosstalk with each other and with DNA methylation. Chromatin immunoprecipitation (ChIP) is the standard assay for choice to analyzing the genomic localization of histone modifications. Exposure to the well-known epigenetic modulator, the DNA methylation inhibiting agent 5-Azacytidine, resulted in a global reduction of DNA methylation in Daphnia magna. Surprisingly, one epigenetic mark, H3K27me3 remains unchanged on the investigated loci. The unchanged response in ChIP was contradictory to a significant gene expression responses and to what was expected of this epigenetic modulator. The present study therefore demonstrates differentiated response of LC-MS/MS, ChIP-PCR and gene expression to 5-Azacytidine exposure when characterizing epigenetic stress response in D. magna. Acknowledgements: funding from the Norwegian Research Council (NRC) project 223268 (CERAD).

**WE297**

Role of microRNAs in the response of the European eel Anguilla anguilla to water pollution

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MicroRNAs (miRNAs) are a class of small non-coding RNA. These 20-24 nucleotides-long sequences associate with the 3'-untranslated region (3'-UTR) of target messenger RNAs (mRNAs), and post-transcriptionally regulate the expression of numerous genes by mediating translational repression or mRNA degradation. In mammals, more than 50% of mRNAs are predicted to be the subject of miRNA-mediated control. One miRNA may regulate hundreds of target mRNAs, and one mRNA may contain multiple binding sites for multiple miRNAs, thus resulting in a complex regulatory network. Although miRNAs are involved in regulation of almost all cellular processes, such as development, growth, apoptosis, immunity and maintenance of tissue-specific function, mechanistic aspects of this regulation are not fully understood. In Human, the aberrant expression of miRNAs has been linked to various diseases and toxic environmental factors such as endocrine disruptors or contaminants, and, in fish, toxicity of environmental pollutants that mainly originate from petrogenic and pyrogenic sources such as combustion of fossil fuels and other organic material. Various PAHs, including benz[a]pyrene (BaP), have been demonstrated to cause a wide range of effects in exposed wildlife, including alterations of immune responses, impaired development and reproduction, as well as mutagenesis and carcinogenesis. Most studies to date, however, have used comparably high exposure concentrations, dietary routes of exposure or intrauterine injection to administer BaP, and knowledge of low-dose effects at concentrations around water solubility (approx. 4 µg/L) is generally limited. This route of exposure, however, must be considered highly relevant in light of the distribution of PAHs even into remote aquatic systems. To bridge this knowledge gap, early-life stages of the fathead minnow (Pimephales promelas) will be exposed to waterborne BaP as a model compound to characterize toxicity pathways that drive the sensitivity of early-life stage-fish to PAHs. Molecular responses at the whole transcriptome, proteome and metabolome level will be investigated at the swim-up stage, and quantitatively correlated with effects on apical growth, survival, development, histopathological, and biochemical endpoints 28 d post-hatch. The data generated within this experiment will help to better understand the relevance of aqueous exposure to BaP specifically, and PAHs in general, and provide important insights into the relevance of molecular responses in early-life stages as early-warning biomarkers for apical outcomes in juvenile and/or adult fish. This study is part of the EcoToxChip project (@ecotoxchip).

**WE298**

Exposure to copper during embryogenesis caused temporary increased tolerance in two subsequent generations in the three-spined stickleback (Gasterosteus aculeatus)

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The sustainability of fish populations in our increasingly polluted environment is critically dependent on their ability to adapt via (epi)genetic mechanisms. Copper is an essential element but when present at high concentrations in the water it can become toxic to aquatic organisms. Recent studies in the UK suggest that copper is the most significant metal pollutant threatening fish in UK freshwater. We conducted a series of copper exposures in stickleback to investigate whether prior exposure can result in altered susceptibility in subsequent generations. Stickleback embryos were exposed to 0.015mg/L copper during early life (0-9dpf), causing ~1.2% mortality, ensuring that selection for a tolerant genotype did not occur. They were then kept under control conditions until sexual maturity. Copper pre-exposed fish were shown to have a significantly higher basal copper tissue burden as adults; and upon re-exposure, they showed a differential response compared to control fish. Mortality curves on F1 embryos revealed that embryos originating from parents who were exposed to copper during embryogenesis were significantly more tolerant to copper when compared to controls. This copper tolerance was shown to be still present in the F2 generation, but not the F3 generation. Our data supports the hypothesis that exposure to low levels of copper during early life has the potential to reduce the susceptibility of a vertebrate model across generations. We hypothesise that the multigenerational tolerant phenotype observed was caused by parental imprinting and/or adaptation to altered copper handling. We measured copper tissue burden in F0 adults, resulting in secondary exposure of F1 embryos and their germ cells which gave rise to the F2 generation.

**Emergence and multidimensional interactions of engineered nanoparticles in toxicity (P)**

**WE299**

Do global warming increase bioaccumulation of copper nanoparticle in...
tilapia? J. Kuo, Kaohsiung Medical University; L. Li. W. Chen, Department of Biomedical Science and Environmental Biology, Kaohsiung Medical University, Kaohsiung

Abstract

Nanomaterial technology was well developed in recent years, and it lead to the nanomaterials accumulation in the aquatic organism. Otherwise, increased atmospheric carbon dioxide causing the phenomenon of global warming. However, less understanding about the effect of warming whether increase the bioaccumulation of copper nanoparticles in freshwater fish. The purpose this study to assess whether warming synergistically increase the bioaccumulation of copper nanoparticles in tilapia (Oreochromis niloticus). Tilapia were randomly exposure to 25 nm of copper nanoparticle (0.3 mg/L) under different temperature (26, 28 and 30°) for periods of 7 days for uptake and 7 days for depuration, to analyze the accumulation and toxicity of copper nanoparticle on muscle. Results showed that the copper accumulation of muscle in the high temperature group was higher than that of control group on day 7 of uptake phase, but there were not significant difference. For one day of depuration phase, the copper accumulation of the 30° group was significantly higher than of 26 and 28° groups (p<0.01). However, they are similar accumulation concentration in the end of depuration period. This study concluded that global warming could increase bioaccumulation of copper nanoparticle in tilapia.

WE300

Environmental mixtures of nanomaterials and chemicals: proposal for a consistent nomenclature of mixture effects in environmental organisms D. Kühnel, S. Classen, T. Knautz, M. Hammers-Wirtz, M. Conicet; C. Jimena, Instituto Nacional de Limnología (CONICET-UNL); S. Ancora, University of Siena / Physical sciences, Earth and environment; N. Bianchi, University of Siena / Department of Physical, Earth and Environmental Sciences; I. Corsi, University of Siena / Physical, Earth and Environmental Sciences. The use of nanotechnology is constantly increasing worldwide and their release into the environment is thus expected, especially in aquatic ecosystems, which are considered the ultimate sink. The use of nano silver (AgNP) based products as antimicrobial agents is undergoing a rapid increase in terms of production due to its biocidal properties. Here we studied the impact of AgNP-based commercial products named NanArgen (Nanotek S.A.) on a common bivalve species, the Mytilus galloprovincialis in terms of biological responses and Ag accumulation. Animals were in vivo exposed for 96h to NanArgen product containing 20-40nm AgNP, according to the manufacturer, at two different concentration (1 μg and 10 μg/L) using natural sea water (NSW) as exposure media. Lysosomal membranes stability (NRRT) and micronuclei frequency (MN) were measured in mussel's hepatocytes. Catalase (CAT) and glutathione-s-transferase (GST) activities were measured in digestive gland as well as the content of malondialdehyde (MDA) and metallothioneins (MT). Effect on multi xenobiotic resistance (MXR) phenotype was assessed by measuring efflux ABC transporters also in vitro using mussel's gills. Total Ag was analyzed in exposure waters after 24h and in mussel's soft tissue after 96h. DLS analysis as well as TEM have been also performed on NanArgen formulation in NSW. A significant increase in lysosomal destabilization and MN frequency was observed in hemocytes of mussels exposed to both concentrations of NanArgen. Furthermore, MT content was significantly higher in digestive gland of mussels exposed only to 10 μg/L while oxidative stress parameters did not show any change compared to controls. A slight negative effect on MXR functionality is observed in vitro. The herein presented effects of nanomaterials in vivo and in vitro contributed to a more comprehensive understanding of mixture effects. The tiered approach results in a consistent terminology to unambiguously describe the different mechanisms of mixture effects that may occur in environmental organisms. Acknowledgement - DK was partially funded by the German Federal Ministry for Education and Research (BMBF) in the frame of the DaNa2.0 project (Data and knowledge on nanomaterials), grant no. 03X0131.

WE301

Investigating the Trojan horse effect of nanoparticles on an aquatic community - An outdoor mesocosm study T. Strauss, Research Institute gaiac; gaiac - Research Institute for Ecosystem Analysis and Assessment; S. Claissen, Research Institute gaiac; T. Knautz, M. Hammers-Wirtz, Research Institute gaiac; gaiac - Research Institute for Ecosystem Analysis

Carbon based manufactured nanomaterials (C-MNMs) are promising materials in nanotechnology. Although both fullerenes and carbon nanotubes have been detected in aquatic organisms, there is a lack of data on their bioaccumulation, potential toxic effect or influence on the food web. In principle, pollutants can become more bioavailable by adsorption to carbon-based nanomaterials. In addition, a spatial transfer of contaminated nanoparticles from the water phase to the sediment could increase the exposure to benthic macroinvertebrates but might also reduce the effect on the planktonic organisms. An outdoor freshwater mesocosm study was conducted with C60 fullerenes and the biocide triclocarban (TCC) using twelve outdoor ponds with a water volume of 3 m³. In addition to uncontaminated controls, both substances were tested alone and in combination. The aim of this mesocosm study is to investigate long-term effects of C60 fullerenes on the community level and to assess their potential to affect the toxicity of TCC. In this outdoor mesocosm study direct and indirect effects on single species as well as on community level endpoints like diversity were evaluated. The taxonomic groups of investigated species were Daphnia, zooplankton (e.g. Daphnia species), and macroinvertebrate species (e.g. chironomids, mayflies, oligochaetes, leeches). Different sampling techniques were used in order to include macroinvertebrates living on and within the sediment as well as hatching insects. In this presentation the results of the mesocosm study will be presented. This work has been supported by the German Federal Ministry of Education and Research (BMBF) as part of the NANO-transfer project.

WE302

Nano silver based products and environmental challenges: toxicity and accumulation in a marine sentinel species G. Liberti, University of Siena / Department of Physical, Earth and Environmental Sciences; A. Ale, Inalı-Conicet; C. Jimena, Instituto Nacional de Limnología (CONICET-UNL); S. Ancora, University of Siena / Physical sciences, Earth and environment; N. Bianchi, University of Siena / Department of Physical, Earth and Environmental Sciences; I. Corsi, University of Siena / Physical, Earth and Environmental Sciences. The use of nanotechnology-based consumer products is constantly increasing worldwide and their release into the environment is thus expected, especially in aquatic ecosystems, which are considered the ultimate sink. The use of nano silver (AgNP) based products as antimicrobial agents is undergoing a rapid increase in terms of production due to its biocidal properties. Here we studied the impact of AgNP-based commercial products named NanArgen (Nanotek S.A.) on a common bivalve species, the Mytilus galloprovincialis in terms of biological responses and Ag accumulation. Animals were in vivo exposed for 96h to NanArgen product containing 20-40nm AgNP, according to the manufacturer, at two different concentration (1 μg and 10 μg/L) using natural sea water (NSW) as exposure media. Lysosomal membranes stability (NRRT) and micronuclei frequency (MN) were measured in mussel's hepatocytes. Catalase (CAT) and glutathione-s-transferase (GST) activities were measured in digestive gland as well as the content of malondialdehyde (MDA) and metallothioneins (MT). Effect on multi xenobiotic resistance (MXR) phenotype was assessed by measuring efflux ABC transporters also in vitro using mussel’s gills. Total Ag was analyzed in exposure waters after 24h and in mussel’s soft tissue after 96h. DLS analysis as well as TEM have been also performed on NanArgen formulation in NSW. A significant increase in lysosomal destabilization and MN frequency was observed in hemocytes of mussels exposed to both concentrations of NanArgen. Furthermore, MT content was significantly higher in digestive gland of mussels exposed only to 10 μg/L while oxidative stress parameters did not show any change compared to controls. A slight negative effect on MXR functionality is observed in vitro. The herein presented effects of nanomaterials in vivo and in vitro contributed to a more comprehensive understanding of mixture effects. The tiered approach results in a consistent terminology to unambiguously describe the different mechanisms of mixture effects that may occur in environmental organisms. Acknowledgement - DK was partially funded by the German Federal Ministry for Education and Research (BMBF) in the frame of the DaNa2.0 project (Data and knowledge on nanomaterials), grant no. 03X0131.
observed reduction in SVL, added to decreased feeding rates, in tadpoles exposed to Au-NR, are important effects that may compromise the fitness of the organisms, since they may cause a delay in the metamorphosis, leading to a longer exposure period of tadpoles to the chemical and to an increase in the time to reach adult and reproductive stage. The higher activity of LDH, at 0.002 μg/mL, may suggest that tadpoles activate first (at low concentrations of Au-NR) a detoxification pathway involving LDH. Furthermore, an enhanced reactive oxygen species (ROS) may have led to the inactivation of catalase and the induction of the oxidative stress-related mechanisms of cell apoptosis. Thus, the induction of GST at the two highest Au-NP concentrations, suggest that the cells are inactivating the Au-NP by its conjugation with reduced glutathione. The results obtained in the present work indicates that Au-NP may induce several sublethal effects in tadpoles of X. laevis that compromise their fitness. Furthermore, since these effects occur at very low concentrations (as low as 0.002μg/mL) it should be classified as “extremely toxic” (EC20 < 0.1 μg/mL; CEC, 1996), suggesting a high environmental risk.

**WE304 Interaction of the biocide triclocarban and weathered multiwalled carbon nanotubes (wMWCNT) in freshwater algae: chronic effects & bioaccumulation**

L. Poliowski, M.P. Hennig, H. Hollert, RWTH Aachen University / Institute for Environmental Research; A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics

The disposal of various plastic nanocomposites containing multiwalled carbon nanotubes (wMWCNT) is increasing in the next decades. Thus, WMCNT release into the aquatic environment due to degradation of the polymers is inevitable. Changes in their properties might happen by several abiotic influences, like weathering by sunlight radiation. WMCNT undergo considerable structural changes compared to their pristine state, like formation of hydroxyl and carboxyl groups on the surface of the WMCNT. As a consequence, weathered WMCNT (wMWCNT) have altered agglomerate- and sorption-sorption properties compared to triclocarban (TCC). This might lead to a different environmental fate of both the wMWCNT and the contaminants in aquatic ecosystems and eventually an enhanced chronic or mixture effect on organisms like freshwater algae. In our studies we examine the chronic effects of wMWCNT and the ‘Troyan horse’ effects of TCC in presence and absence of wMWCNT on Pseudokirchneriella subcapitata and Chlamydomonas reinhardtii. Growth inhibition tests were performed according to OECD test guideline 201. In a first part of experiments, the growth inhibition of both species was tested in the range of 10 – 60 μg/L for TCC and 0.1 – 16.0 mg/L for wMWCNT. The mixture toxicity of a TCC test series (10 – 60 μg/L) and 100 μg/L wMWCNT was additionally investigated on P. subcapitata. A second series of experiments was carried out by adding the highest toxicity (60 μg/L) to variable wMWCNT concentrations to figure out, which wMWCNT amounts are necessary to reduce the toxicity of TCC. We determined a concentration dependent growth inhibition of P. subcapitata for TCC and TCC + 100 μg wMWCNT/L with an EC50 of 37 and 36 μg TCC/L, respectively. This amount of wMWCNT appears to be not sufficient to adsorb the entire free TCC from the water phase, which eventually (wMWCNT > 50 μg/L) may lead to very similar EC50 values in both scenarios. Only in a second experiment it could be shown, that wMWCNT amounts > 1 mg/L reduce algae growth inhibition completely causing 60 μg TCC/L. Regarding to mixture effects of TCC and wMWCNT to algae, bioaccumulation of wMWCNTs by freshwater algae needs to be investigated, especially in respect to long term incubation times and low wMWCNT amounts. **Acknowledgements** The work is supported by the European Powder-2-Transfer that receives funding from the Bundesministerium für Bildung und Forschung (BMBF) under agreement with the FP7 ERA-NET SIENN.

**WE305 Comparative assessment of the interactive effects of Carbon-based nanomaterials and Benzo(a)pyrene on zebrafish embryos**

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This study aimed to assess the toxicological consequences related to the interaction of Benzo(a)pyrene (B(a)P) with two CNMs with different physico-chemical properties, namely carbon nanopowder (CNPW) and fullerene (C60), performed in this aim to determine the effective sorption of the hydrocarbon on CNMs was quantified. A thorough evaluation of chemico-physical interactions between the two CNMs and B(a)P has been performed. Embryos were exposed to CNPW, C60 and B(a)P alone and their combination. The uptake of CNMs and B(a)P and their localization in embryos were assessed by immunofluorescence and electron microscopy. To evaluate the toxic effects of interaction of B(a)P with CNMs, a set of biomarkers of genotoxicity and oxidative stress was applied. Proteomics analysis allowed also the identification of molecular events involved in the responses to pollutants alone and in co-exposure. Overall results showed that the different physico-chemical properties of the two CNMs influenced their interactions with B(a)P and generated distinct toxic effects. Indeed the adsorption on CNPW modified the accumulation of B(a)P, which followed the distribution of the physical pollutant instead of its natural bioaccumulation. On the contrary the co-exposure with C60 did not affect the uptake/distribution of B(a)P. Instead, C60 doped with B(a)P is more prone to sedimentation and less bioavailable for the embryos compared to C60 alone. The integrated results from biomarkers and proteinomics showed that different stress responses were activated by the different sorption of carbon nanoparticles in combination with B(a)P. The CNPW doped with B(a)P mainly mirrored the effects shown by the physical contaminant rather than by the hydrocarbon, while C60 doped with B(a)P seems to induce a cellular response similar to B(a)P alone. The study highlighted that in the aquatic ecosystems complex interactions are established between pollutants and CNMs which could elicit unexpected ecotoxicological effects.

**WE306 IN VITRO TOXICITY OF MODEL ZnO NANO Particles on Hemocytes of MusSEL Mytilus gallopoRvinicisL.**

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Zinc oxide nanoparticles (ZnO NPs, size 58 nm, as calculated using X-Ray diffraction data) were manufactured through Flame Spray Pyrolysis, and their effects were subsequently investigated on hemocytes of mussels Mytilus gallopoRvinicisL. Following the collection and preparation of cell suspensions, mussel hemocytes were treated for 1 h with different concentrations of ZnO-NPs (5, 10, 25 and 50 μg/mL), dispersed with a probe sonicator, as well as ZnCl2 (10 and 25 μg/mL); positive control, from a stock solution of ZnCl2, in 2dH2O, in 1:1 ratio. Afterwards, stress indices such as (a) cell viability (in terms of Neutral Red Retention Time/NRRT assay) (b) the generation of superoxide radicals (O2·-), using Nitro blue tetrazolium/NBT, (c) the production of nitrogen oxides (NO, in terms of nitrites), and (d) lipid peroxidation (in terms of malondialdehyde/MDA equivalents) were measured. The results demonstrated a significant increase of cell death after treatment with ZnO NPs at concentrations higher than 5 μg/mL, with maximum values (>50%) of cell death after exposure to ZnO NPs 50 μg/mL. Furthermore, hemocytes treated with sub-lethal concentrations of ZnO-NPs (5-25 μg/mL), showed a significant increase of O2·- and NO and MDA, compared to those values observed in control cells in each case. Finally, the results of the exposure to ZnO NPs were compared with the respective results after exposure to ZnCl2, showing a similar pattern. Those effects of ZnO NPs on mussel hemocytes confirm the cytotoxic and oxidative potential of well-promised nanomaterials, such as ZnO NPs, widely used in a variety of new cutting-edge applications.

**WE307 Toxico-transcriptomics as tool to identify nano-specific toxicity profiles**

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The use of omics is rapidly increasing in the field of nano-ecotoxicology; an increasing number of studies are aiming to investigate the effects and mode of action of engineered nanomaterials (ENM) in this way. However, a systematic synthesis of the outcome of these studies in order to identify common responses between ENM and organism groups has not yet been performed. We therefore established a computational analysis pipeline with the aim to re-analyze various transcriptomic datasets in a consistent manner. The pipeline allows a re-mapping of array probe sequences, followed by established statistical analysis and thus improves data set comparability. Differentially expressed genes (DEGs) are determined by comparison between treatment and untreated samples (qFDR)

**WE308 Zinc toxicity to A549 cells and Daphnia magna changes with iron oxidation nanoparticles**

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The use of iron oxide nanoparticles (IONPs) as an environment remediation tool is based on their ability to adsorb and immobilize metals and decrease their bioavailability. However, the adsorption of metal contaminants by nanoparticles can also potentially increase the toxicity of either NPs or the metals, for example due the Trojan horse effect. In this study, we evaluated the acute effect of zinc (Zn) as zinc sulfate heptahydrate (ZnSO4·7H2O) after an incubation period with a fixed concentration of humic acid (ha) coated IONPs (ha-IONPs), on the in vitro toxicity to human A549 cells and on the toxicity to Daphnia magna as a model freshwater invertebrate species. Non-toxic concentrations of ha-IONPs were selected for the
assays taking also into account the predicted adsorption of Zn. The ha-IONPs concentrations used were 0.45g/L and 0.52 g/L for the A549 and the Daphnia magna experiments, respectively. In A549 cells, the incubation of Zn with ha-IONPs did not change the Zn effects on cell viability after 24h in terms of IC_{50} (0.006 g/L vs. 0.010 g/L with and without ha-IONPs, respectively). However, the shape of the dose-response curve became shallower (e.g., the IC_{50} for Zn was 0.070 g/L with and without ha-IONPs, respectively). This indicates a potential protective effect of IONPs at high metal concentrations and a synergistic effect at low metal concentrations. These experiments were also conducted in the presence of serum proteins, and despite the toxicity of Zn decreased, the same effect of co-incubation with ha-IONPs was observed. Optical micros cope images showed that ha-IONPs aggregates were uptake by the cells during the experiments. Therefore, even if adsorbed on ha-IONPs, NMs would reach intracellular compartments. Differences in the relative uptake of free vs. ha-IONP adsorbed zinc as well as intracellular bioavailability of Zn in these two forms would be explaining the changes in the dose-response curve that were observed. Acute studies (up to 48-hours) with Daphnia magna showed a protective effect of the ha-IONPs on the toxicity of Zn. The EC_{50} value for Zn increased from 0.23 mg Zn/L to 1.1 mg Zn/L in presence of ha-IONP. According to DLS data, the adsorption of Zn to NPs decreased their stability and subsequently increased their co-precipitation in the exposure media. This settling process would decrease the bioavailable zinc concentration in the exposure medium and therefore its toxicity in Daphnia magna.

WE309 Internalization of graphene-related nanomaterials in fish cell lines J. Kalman, Instituto Nacional de Investigación y Tecnología Agraria y Alimentaria / Environment; C. Merino, Grupo Antolin Ingenieria SA; M. Fernandez-Cruz, J. Navas, INIA - National Institute for Agricultural and Food Research and Technology / Environment

The H2020 NanoReg2 project aims to develop and implement grouping and Safe by Design strategies for nanomaterials (NMs). For this purpose, toxicity of selected carbon-based NMs have been assessed in order to refine Safe by Design approaches considering three pillars: safer product, safer use, and safer production process. Graphene-related nanomaterials (GRMs) are among the newest and most important NMs. Their extraordinary physicochemical properties have attracted great interest in most areas of science and industry. Nevertheless, the incorporation of these NMs into products inevitably leads to their release into the aquatic environments. In a previous study we assessed the cytotoxicity of tubular-shaped carbon nanofibers (CNFs) and graphene oxide (GO) on fish hepatocytes (derived from topminnow fish, Poylophisius lucidu) and macrophages (derived from carp leukocytes, Cyprinus carpio). In general, the observed IC50 values after 72h exposure were higher than 100 µg/ml with some exceptions in the case of CLC cells. In the present work we focussed on the uptake and intracellular fate of these NMs. Cells were exposed to three different concentrations (non-toxic, low toxic and relatively toxic) of each NM for 72 h. Transmission electron microscopy was used to investigate possible internalization and intracellular fate of these NMs in hepatocytes and macrophages. All GRMs were visualized in both cells even at the lowest exposure concentrations. Carbon nanofibers were taken up into vesicles of hepatocyte cells in a size-independent manner, whereas in macrophages, longer CNFs were encountered free in the cytoplasm and only the shorter CNFs were localized in membrane-bound compartments. GO sheets were present within vesicles as well as free in the cytoplasm of both cell types. Understanding the behaviour of these NMs in aquatic systems is a key issue for safe handling of these NMs. This research is supported by the EU’s Horizon 2020 research and innovation programmes (NanoReg2, Grant Agreement nº 646221 and MSCA-IF-2016, Grant Agreement nº 746876).

WE310 Molecular mechanism and physicochemical properties of Cadmium-TiO2 nanoparticle mixtures when co-exposed to the nematode Caenorhabditis elegans L. Kleene, Hamburg University of Applied Sciences (HAW) / Life Sciences; A. Hursthouse, University of the West of Scotland / School of Science; S. Heise, Hamburg University of Applied Sciences / Life Sciences.

The number of engineered nanomaterials (ENM) is rising continuously in consumer care products, from sunscreens alone into commercial products. About 93% of ZnO-NP-containing products are paints, cleaning and personal-care products, from which ZnO-NP can be easily leached. Annually, around 250 tonnes of ZnO-NP were estimated to be released from sunscreens alone into the marine environment. However, there are no comprehensive regulations of NMs, including ZnO-NP, in any countries due to the diverse physicochemical properties of NMs and their complicated interactions with the environment. In the marine environment, ZnO-NPs are exposed to various environmental factors, such as temperature and salinity, but influences of these factors on the physicochemical properties and toxicity of ZnO-NP are often tested individually. As both factors co-exist in the environment and may influence one another, it is vital to study their effects concurrently to tease out any potential interactions. This study, therefore, investigates the interacting effects of temperature and salinity on ZnO-NP to a common marine copepod (Tigriopus japonicus) along the Western Pacific coast. Physicochemical properties (aggregate size & ion dissolution) of three zinc-associated compounds, including ZnO-NP, ZnO bulk-particles (ZnO-Bulk) and ZnSO4·H2O (ZnSO4), were characterized to compare their toxic mechanisms. Acute toxicity was determined with 96-h standard toxicity tests under nine different combinations of temperature (15, 25 & 35 °C) and salinity (12, 22 & 32 PSU), i.e., a 3 x 3 factorial design. Preliminary results showed that increase in temperature and salinity could increase aggregate size of ZnO-NP and ZnO-Bulk, but reduce their ion dissolution rate. At 25 °C, similar to previous studies which suggest that ZnO-NP has a higher toxicity with temperature compared to ZnO-Bulk, acute exposure to 22 PSU, where the dissolution rate of Zn2+ was the smallest, ZnSO4 was the least toxic compound, implying that Zn2+ were not the only contributor to the observed toxicity. Higher toxicity of ZnO-NP and ZnO-Bulk might be due to particle and animal interactions such as physical damage by larger aggregate at larger salinity. The results will advance our understanding of interactions of ZnO-NP and ZnO-Bulk with aquatic organisms, and be able to determine effect threshold concentrations for regulation of products of ZnO-NP under different combinations of these two environmental factors.

WE312 Multigenerational effects of gold nanorods to Raphidocelis subcapitata and Chlorella vulgaris C. Monteiro, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro; C. Venancio, Department of Biology / Biology; A.L. Daniel-da-Silva, S.F. Soares, University of Aveiro / Department of Chemistry & CICECO, 3810-193 Aveiro; A.M. Soares, University of Aveiro / department of Biology & CESAM; T. Trindade, University of Aveiro / Department of Chemistry & CICECO; L. Lopes, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro

Multigenerational effects of gold nanorods to Raphidocelis subcapitata and Chlorella vulgaris. C. Monteiro, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro. The multigenerational effects of gold nanorods (Au NPs) to the freshwater microalgae Raphidocelis subcapitata and Chlorella vulgaris were estimated to be released from sunscreens alone into the marine environment. However, there are no comprehensive regulations of NMs, including ZnO-NP, in any countries due to the diverse physicochemical properties of NMs and their complicated interactions with the environment. In the marine environment, ZnO-NPs are exposed to various environmental factors, such as temperature and salinity, but influences of these factors on the physicochemical properties and toxicity of ZnO-NP are often tested individually. As both factors co-exist in the environment and may influence one another, it is vital to study their effects concurrently to tease out any potential interactions. This study, therefore, investigates the interacting effects of temperature and salinity on ZnO-NP to a common marine copepod (Tigriopus japonicus) along the Western Pacific coast. Physicochemical properties (aggregate size & ion dissolution) of three zinc-associated compounds, including ZnO-NP, ZnO bulk-particles (ZnO-Bulk) and ZnSO4·H2O (ZnSO4), were characterized to compare their toxic mechanisms. Acute toxicity was determined with 96-h standard toxicity tests under nine different combinations of temperature (15, 25 & 35 °C) and salinity (12, 22 & 32 PSU), i.e., a 3 x 3 factorial design. Preliminary results showed that increase in temperature and salinity could increase aggregate size of ZnO-NP and ZnO-Bulk, but reduce their ion dissolution rate. At 25 °C, similar to previous studies which suggest that ZnO-NP has a higher toxicity with temperature compared to ZnO-Bulk, acute exposure to 22 PSU, where the dissolution rate of Zn2+ was the smallest, ZnSO4 was the least toxic compound, implying that Zn2+ were not the only contributor to the observed toxicity. Higher toxicity of ZnO-NP and ZnO-Bulk might be due to particle and animal interactions such as physical damage by larger aggregate at larger salinity. The results will advance our understanding of interactions of ZnO-NP and ZnO-Bulk with aquatic organisms, and be able to determine effect threshold concentrations for regulation of products of ZnO-NP under different combinations of these two environmental factors.
cetyltrimethylammonium bromide (CTAB) was quantified by exposing both algae to the concentration of CTAB present in the highest tested concentration of Au-NP: 90 µg/L for C. vulgaris corresponding to 0.257 nM of CTAB. Chlorella vulgaris exhibited a higher tolerance to Au-NP than R. subcapitata: EC50 for F0 was 79 µg/L and 39 µg/L, respectively. For C. vulgaris, a gradual increase of its tolerance to Au-NP was observed over generations; after being exposed for four generations to this chemical, the effect was significant on growth rate were inhibited among all concentrations and the control. A different pattern of response was observed for R. subcapitata. This species significantly increased its sensitivity to Au-NP from F0 to F2 generation, but recovered a similar sensitivity to that quantified for F0 from F2 to F4. CTAB significantly reduced the growth of microalgae by 42%. Over generations, the sensitivity of the two algae species to CTAB was not changed. The results obtained in the present work reveal that traditional standard assays with short-term exposure may over- or underestimate the real risk posed by Au-NP to freshwater microalgal. Therefore, it is suggested that long-term exposures should be included in the ecological risk assessment.

WE313 Effects of climate change combined with copper nanoparticle on early development of Japanese medaka (Oryzias latipes)

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Nowadays, global warming and acidification were occurred by rising carbon dioxide (CO2). The factory have been continuously emit copper nanoparticle into ocean and river. They probably induced harmful biological effect on organisms. However, the combined effects of three environmental stressors on aquatic species have not been well studied. The purpose of this study is to assess the effects of aquacultural acidification and warming combined with copper nanoparticle on survival and hatchability of early development of Japanese medaka (Oryzias latipes). Firstly, the embryos were exposed to 25 nm copper nanoparticle (30 µg/L) and without copper nanoparticle under nine temperature and pH conditions (26°C/7.5, 26°C/6.5, 26°C/5.5, 28°C/7.5, 28°C/6.5, 26°C/5.5, 30°C/7.5, 30°C/6.5 and 30°C/5.5) for 14 days to observe hatchability and mortality. Then, the newly hatched fry were exposed to same condition for 14 days to observe the survival. Results showed that the mortality of embryos had a upward trend, and the hatching rate had a downward trend in exposure group of pH 5.5 compared with that of others pH groups. However, only 28°C/5.5 combined with copper nanoparticle group was significantly increased mortality and inhibited hatchability than that of 28°C/6.5 combined with copper nanoparticle group. While the survival rate of larvae had a significantly downward trend in exposure groups of lowest pH level and highest temperature on days 10. In conclusions, medaka larvae have more sensitive than embryos by survival ability. Moreover, aquatic acidification and warming were synergistic effect to increase copper nanoparticle toxicity to larvae. Therefore, we found that three environmental stressors caused a potential hazards to fish population.

WE314 The use of the marine mussels Mytilus hemocyes as a model for studying the impact of NPs on innate immunity

M. Auguste, University of Genova / DISTAV; T. Balbi, L. Canesi, university of genou / DISTAV

Nanoparticles (NPs) are widespread used in consumer products and industry; they are getting more interesting interaction with human and also their potential impact on environmental species. Recent studies have demonstrated that NPs could affect a number of marine species, and interact with their immune system. Within the PANDORA project (Probing safety of nano-objects by defining immune responses of environmental organisms), a European Training Network (ETN) funded in the framework of H2020 Marie Sklodowska-Curie ITN programme, objectives are to identify immunological mechanisms triggered by nano-objects, and predictive markers of risk vs. safety, with a collaborative cross-species comparison. The use of mussel hemocytes, from Mytilus galloprovincialis, for in vitro testing is a valuable tool in the screening of the toxicity of NPs as the tests are inexpensive and use alternative animals in experimental studies. In this study, we analyzed the impact of colloidal immune parameters (e.g. lysosomal membrane stability, superoxide and NO production, phagocytic activity) and particle internalisation by hemocyte upon short-term exposure to NPs (different concentrations and times of exposure from 30 min to 1h). Once entering the organism, NPs are in contact with other type of media e.g. hemolymph serum for mussels. For some type of NPs, the response is affected by adsorption of protein components of hemolymph serum involved in the formation of a NP-protein corona. In order to have a wider view of the interactions and mechanisms of actions of NPs, the same parameters are measured with NPs suspensions in artificial seawater (ASW) and serum. The results obtained with Mytilus hemocytes will be compared with those obtained in immune cells of other model organisms within the PANDORA project. According to the special properties of every NPs, the aim is to understand the main mode of action at the cell level that will help designing predictive in vitro assays to measure the immuno-risk of NPs to the environment in the future. *Funded within the European Union’s Horizon 2020 research and innovation programme under the Marie Sklodowska-Curie grant agreement PANDORA No 671881.

WE315 Influence of warming and acidification on copper nanoparticle bioaccumulation in medaka (Oryzias latipes) embry

Y. Zhang, L. Meng, L. Yan, Department of Biomedical Science and Environmental Biology, Kaohsiung Medical University, Kaohsiung; M. Yallop, G. Barker, University of Bristol

The development of industry and technology in the last decade has increased the production engineered nanomaterials (ENMs) used widely in fields such as medicine, energy, agriculture and consumer goods production. As a result, there are increasing concerns about the release of engineered nanomaterials such as metal oxide nanoparticles into the environment. While the effect of metals and ENMs as single contaminants have been extensively studied, much research is needed to account for potential mixture effects due to heavy metal-ENMs interactions. Studies suggest that the toxicity of ENMs such as metal oxide nanoparticles is mainly due to the release of dissolved metal ions. However, majority of these studies have mainly focused on impact of ENMs in freshwater environment and results are extrapolated for other types of environmental systems (marine, soil, sediment). Evidence is accumulating that the dissolution of ENMs is dependent on mineralogy and the conditions of the exposure medium. In this research, the combined toxicity of heavy metals (Cu and Zn) and their metallic oxide nanoparticles will be evaluated in a benthic estuarine microalgal - Cylindrotheca closterium. We hypothesize that the aggregates formed by the metal-oxide nanoparticles may adsorb the free metal ions, reducing the bioavailable fraction of the metal and thus alter potential bioavailability and toxicity of metal-ENMs interactions.

WE316 Assessing the combined toxicity of metals and metal-oxide nanoparticles in a benthic estuarine microalgal

R.B. Ogunjumobi, M. Yallop, G. Barker, University of Bristol

Coastal aquatic ecosystems such as estuaries are at risk of metal pollution due to anthropogenic inputs from acid mine drainage, industrial and agricultural run-off. In addition, advances in nanotechnology in the last decade have increased the anthropogenic inputs from acid mine drainage, industry and agriculture. Hence, environmental fate and unintentional ecological effects and/or toxicities of nanomaterials and massively used for multiple purpose to improve human life. In addition, nanotechnology in the last decade has increased the environmental fate and unintentional ecological effects and/or toxicities of nanomaterials and massively used for multiple purpose to improve human life. Therefore, we concluded that there was no influence of Cu accumulation in medaka embryo when warming and acidification occurs in the future.

WE317 Comparative toxicity of silver nanocolloids and titanium dioxide nanoparticles using medaka

Y. Kato, Toyo University / Faculty of Life Science; T. Ariyoshi, C. Kataoka, S. Kashiwada, Toyo University / Graduate School of Life Sciences

Silver nanoparticles and titanium dioxide nanoparticles are representative nanomaterials and mass-use semiconductor that multi purpose to improve human life. Hence, environmental fate and unintentional ecological effects and/or toxicities have been concerned and many studies are reported using model organisms. We have been investigating fish toxicity and ecological risk of silver nanocolloids (SNCs, 40 nm) using medaka model. SNCS have embryonic (at 0.5 mg/L of SNC) and larvae (at 5 mg/L of SNC) toxicities including lethality, inhibition of embryo development, shortening of length, small eye development, ischemia, reduced heart beating, and caused some oxidative stresses such as GSH reduction and lipid peroxidation. To adults, SNCs exposure (at 5 mg/L) did not exhibit significant lethality; however, it was observed that SNCs exposure (at 0.05 mg/L) disrupted immune system and reduced tolerance to infective bacterial disease ( Edwardsiella tarda). In SNCs exposure, we resulted that silver chloro-complexes, which were made of dissociated silver ion from SNCS, should be essential toxicants of SNCS exposure. On the other hand, titanium dioxide nanoparticles (TiO2-NP, Φ < 90 nm) are well-known causing oxidative stress by UV radiation; however, there are some reports that TiO2-NP does not have significant toxic effect to fish other than
hypertrophy of gill nubb. We have assumed that ion dissociation will be a key to understand nano-toxicity depended on materials. TiO₂-NP which does not dissociate ions, was employed as a reference to ion dissociation NP (i.e. SNCs). In exposure of TiO₂-NP (at 10 mg/L) to embryo and larvae, there was no biological toxic effect mentioned above at all. In our presentation, we will discuss comparative toxicity of SNCs and TiO₂-NP regarding general toxicity, oxidative stress, cytotoxicity (apoptosis and necrosis), immunotoxicity, and tolerance to reactive biohumoral diabetic disease. Through this study, we will figure out that dissociated ions should be toxic essential of nanomaterials, and not always nano-sized material will be toxicant in aquatic environment.

WE318 Genotoxic assessment of aluminium oxide nanoparticles in relation to Escherichia coli and Aeromonas hydrophila n. doskocz, M. Zalęska-Radziwill, A. Affek, Warsaw University of Technology, Faculty of Building Services, Hydro and Environmental Engineering / Department of Biology

Increasing production and use of nanoparticles contributes to their widespread dissemination in the environment and their unique physicochemical properties lead to unlimited distribution in environmental compartments. Migration ability of nanoparticles can have very dangerous consequences, as they can be transferred to potable water. Genotoxicity biomarkers are regarded as useful tools for the assessment of chemical hazards in aquatic ecosystems, because chemicals which damage DNA can significantly alter the functioning of ecosystems. Recent advances in molecular biology have led to the development of several techniques, which can be used for DNA analysis in the field of genetic toxicology. The randomly amplified polymorphic DNA (RAPD) method is a PCR-based technique that amplifies random DNA fragments with the use of single short primers of arbitrary nucleotide sequence under low annealing conditions. RAPD-PCR test has been successfully used for detection of genetic damage in animals and plants. The SOS-Chromotest is one of the most commonly used bacterial tests. It is based on the induction, by genotoxic compounds, of a bacterial SOS repair system conjugated to the β-galactosidase gene and the subsequent measurement of the enzyme expression. In this work, genotoxicity studies on the basis of the RAPD-PCR and SOS-Chromotest assay were performed for aluminium oxide nanoparticles (nAl₂O₃). Lettuce, as zebra fish, on the fate, transport, and effects of nanomaterials, including metal based particles such as nano-Al₂O₃, in the environment. The interest in nano-Al₂O₃ is due to the fact that their influence on genetic material of bacteria is practically unknown. Results obtained for the nanocompound were compared with those for Al₂O₃ macro form. The nanocompound caused changes in the genetic material of bacteria A. hydrophila. Disregard of genetic mobility of obtained profiles bands for primer O2a differed from the results obtained for the negative control by more than 27.3%, while from positive control - only by 15.6%. Furthermore, the largest decrease in genetic stability was 89.3%. The values of genotoxicity induction coefficient (I) in the SOS-Chromotest showed strong genotoxicity for nano-Al₂O₃, in the presence of S9 fraction and slight genotoxicity in the absence of S9 fraction in mutants of E. coli. The results showed also that nano-Al₂O₃ can induce genotoxicity a greater extent than the same compounds in their macro form.

WE319 Effects of Copper Oxide Nanoparticles and Arsenic on the Whole-Life Cycle of Rice (Oryza sativa japonica) I. M. Simms, G. P. Cobb, Baylor University / Department of Environmental Science

Copper oxide nanoparticles (nCuO) and arsenic (As) phytotoxicity to rice plants (Oryza sativa japonica) was evaluated in a factorial study using (0, 0.1, 1.0, 10, 50, and 100 mg/L) in daily watering and As (0 and 10 mg/kg) in soil. Experiments were conducted in a greenhouse during 130 day interval. Two rice plants were grown in 3L plastic pots without drainage. Toxins were quantified in soils and water by inductively coupled plasma-mass spectrometry before, during and after testing. nCuO particle sizes were determined by differential light scattering before addition to the test system. Exposures began at planting, continued through germination, and to seed production. Thus, our study is the first to examine the influence of nCuO in conjunction with As throughout the whole life cycle of rice plants. nCuO significantly affected the seed germination, but both nCuO and As had a significant main effects on the fresh weight (FW) of rice straw and the number of rice panicles (NRP). The interaction of the two toxicants was also significant on both FW and NRP. A decrease in the FW was observed for rice exposed to As, only. The midlevel concentrations (1.0 and 10 mg/L) of nCuO decreased the FW with no As addition, while higher concentrations (50 and 100 mg/L) significantly increased the NRP. Low and high nCuO concentrations (0.1, 5.0, and 100 mg/L) decreased the FW with the As addition, and NRP decreased in treatments receiving As along with either lower or higher nCuO concentrations. More data from this greenhouse study are currently being compiled and analyzed to determine the effect of As and nCuO on grain yield as well as Cu and As uptake, distribution, and speciation in rice plants and the grain.

WE320 Behavior of cerium oxide nanoparticles in presence of pharmaceuticals compounds on aquatic specimens G. AMARIEI, Universidad de Alcalá; K. Boltes, Universidad de Alcalá / Chemical Engineering; P. Letón, Universidad de Alcalá

Nanoparticles, in particular metal oxide nanoparticles, have found extensive usage in a wide range of services and industries. Subsequently, they can be released into environment and finally end up in water bodies. That may suppose a potential risk for aquatic environment, exerting toxic effects at the level of cells/tissues or the whole organisms.² The present study, evaluate the toxicity behavior of cerium oxide nanoparticles (CeO₂-NPs) on three aquatic specimens- algae Selenastrum capricornutum, bacteria Vibrio fischeri, and activated suldge, by exploring concentration-dependent effect and changes induced due to the presence of Ibuprofen (Ibu) or Levofloxacin (Levo). 72h algae growth-inhibition, marine bacteria- luminiscence reduction and 24h sludge enzymatic activity and oxidative stress were used as endpoints. Nanoparticles concentration ranged from 0.6 to 160 mgL⁻¹. The particle size and the ζ-potential of NPs in the culture media were measured to analyse the relation between stability profile and the observed toxicity behavior. The obtained results reveal toxic effects of CeO₂ nanoparticles leading to growth inhibition in algae. The presence of Ibu did not produce significant changes, while Levo showed drastic negative effect in algae growth. Short-term exposure produced significant reduction of luminescence intensity in marine bacteria. The presence of both, Ibu or Levo, reduced the negative effects of single nanoparticles in Vibrio fischeri. Exposure produced significant oxidative stress in bacteria forming activated sludge, with lower damage to enzymatic activity. The presence of pharmaceuticals compounds did not produce significant changes on nanoparticle (n) toxicity to food: 1) macroalgae, 2) bacteria. Results indicated that algae was more strongly affected than the marine bacteria and activated sludge, respectively. These can be attributed to the culture media and organisms structural characteristics, respectively. [1] Neale PA, Jamting AK, O’Malley E, Herrmann J, Escher BL. 2015. Behaviour of titanium dioxide and zinc oxide nanoparticles in the presence of wastewater-derived organic matter and implications for algal toxicity. Environmental Science: Nano-1:193-202. [2] Salgueiro ML, Demonce H, Am Z, Bill H, Heidi G. 2016. Interaction of engineered nanomaterials with hydrophobic organic pollutants. Nanotechnology 27:284003. Acknowledgement - The research was funded by the Comunidad de Madrid, grants S2013/MAY_2716 REMTAVARES.

WE321 Toxicity of nanoparticles of titanium dioxide to Daphnia longispina: waterborne versus dietary exposure E. Padilla, Institute for Environmental Sciences / University Koblenz-Landau; C. Venancio, Department of Biology / I. Lopes, University of Aveiro / Department of Biology / I. Santos, University of Aveiro / Department of Biology / N. Nascimento, University of Aveiro / Department of Biology / REMTAVARES.

Nanoparticles of TiO₂ (nTiO₂) are extensively used in many commercial products. Maybe for this reason, this nanoparticle is amongst the most studied in ecotoxicology. This study intended to discriminate the toxicity caused by n-TiO₂ to the daphnid species Daphnia longispina, either through waterborne or dietary exposure routes. For this, neonates of D. longispina were exposed to a control and to the following n-TiO₂ for 21 days: 1) pure water, 2) with a concentration of 0.625 mgL⁻¹ n-TiO₂ (corresponding to the EC₅₀ for D. longispina), ii) food (microalgae) spiked with nano-TiO₂ (after being exposed for 3 days to a concentration of 0.615 mgL⁻¹ n-TiO₂), and iii) water and food spiked with n-TiO₂. The effects of the nanoparticle were monitored on the feeding rate (after a 24h exposure period), somatic growth rate (after a 10-day exposure) and in reproduction (after 21-day of exposure). short-term (21 days) of D. longispina was exposed to a control and to different doses of n-TiO₂. Results showed that the positive controls had normal growth and reproduction whereas the negative control did not. Exposure to n-TiO₂ caused a significant decrease in several endpoints, including survival, growth rate, and reproduction. The obtained results revealed significant differences in the survival, growth rate, and reproduction of D. longispina between the control and the nano-TiO₂ treatment groups. These findings highlight the potential risks associated with the use of titanium dioxide nanoparticles in aquatic environments.

WE322 Dynamics of Cu accumulation in charophyte cell compartments after its exposure to nCuO S. Manu, M. Manu, S. Manu, Nature Research Centre / Institute of Botany, Laboratory of Aquatic Ecotoxicology; B. Gylyte, S. JURKONIENE, R. Vitkus, Nature Research Centre / Institute of Botany

In plant and bacterial cells, prior to be internalized, NPs have to pass semipermeable cell wall and plasma membrane barrier. Metal nanoparticles may interact with the cell directly or induce toxicity through the release of metal ions. Internalized cell of characean green alga poses features such as big size and separation of the main cell compartments, which enable mechanical separation of the compartments, namely cell wall, cytoplasm and vacuole. In this study, fractionation procedure was verified by cytoplasm and vacuole specific biomarkers.
malate dehydrogenase and α-Mannosidase, respectively. A high-purity vacuumolar (99.5%) and cytoplasm (86.7%) fractions of the cells of *Nitzschia obtusa* were obtained. The cell wall fraction contained approximately 1.8 and 13.4% of cytoplasm and vacuole. By additional washing of the cell wall it was possible to diminish contamination with cytoplasm. The data on Cu accumulation dynamics within the compartments after cell exposure to rCuO suspensions will be presented and the role of the cell wall in the accumulation process will be discussed.

**WE323** Are graphene nanomaterials "Trojan horse" carriers for oil compounds in mussel hemocytes in vitro?

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In the aquatic environment, complex mixtures of pollutants are usually found. Polycyclic aromatic hydrocarbons (PAHs) are priority pollutants and main constituents of the water accommodated fraction (WAF) of petroleum. Graphene nanoplatelets can adsorb organic compounds thus being potentially useful in oil spill remediation. However, they could also act as vehicles of organic contaminants to aquatic organisms ("Trojan horse" effect). This study aimed to evaluate the "Trojan horse" carrier effect of graphene nanoplatelets. This work was funded by the EU H2020 GRACE project (grant 679266), Spanish MINECO (project NACE, CTM2016-81330-R), Basque Government (consolidated research group IT10-13) and University of the Basque Country (UIF 11/37).

**WE324** Multigenerational effects of titanium dioxide and silver nanoparticles on *Daphnia magna*: gene expression and morphological changes in the presence or absence of aged nanomaterials

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Recent studies have investigated nanoparticle (NP) physicochemical properties and interactions with biological systems. *Daphnia magna* was chosen as the model organism, as they are well characterized and reproduce parthenogenetically, which is well suited for experimental genetic studies and monitoring stress/adaptive responses. The current research suggests that sediment organic compounds (graphene oxide (GO), GO-polyvinylpyrrolidone (GO-PVP) and reduced GO-PVP (rGO-PVP)) with adsorbed oil compounds from naphthenic North Sea crude oil WAF using *in vitro* toxicity assays in hemocytes of marine mussels. Two approaches were tested to obtain graphene nanoplatelets with adsorbed oil compounds: filtration and centrifugation. Hemocytes were exposed to a wide range of concentrations of GO, GO-PVP and GO-PVP with and without adsorbed oil compounds and to a series of WAF dilutions. After 24 h exposure, cell viability (MTT assay) and ROS production were assessed. Centrifugation (270g for 30 min) successfully separated WAF solution from graphene nanoplatelets with adsorbed oil compounds. This procedure was thus used for *in vitro* toxicity testing. WAF decreased cell viability and increased ROS production in hemocytes starting at 25% WAF. GO, GO-PVP and rGO-PVP nanoplatelets were moderately toxic to mussel hemocytes and produced a significant increase in ROS production. In exposures to graphene with adsorbed oil compounds, hemocytes viability decreased at similar concentrations as in exposures to nanoplatelets alone. However, ROS production increased in hemocytes exposed to lower concentrations of graphene with adsorbed oil compounds (10 ML/L) compared to nanoplatelets alone (25 ML/L), indicating that adsorbed oil compounds increase nanoplatelets toxicity. In conclusion, a protocol to obtain graphene nanoplatelets with adsorbed oil compounds was established. Nanoplatelets with and without adsorbed oil compounds showed similar cytotoxicity to hemocytes but the ones with adsorbed oil compounds increased ROS production earlier, indicating that graphene nanoplatelets may act as "Trojan horse" carriers of oil compounds. This work was funded by the EU H2020 GRACE project (grant 679266), Spanish MINECO (project NACE, CTM2016-81330-R), Basque Government (consolidated research group IT10-13) and University of the Basque Country (UIF 11/37).

**WE325** Evaluating the contribution of environmental stressors to sediment concentrations of PAHs in the northern Gulf of Mexico

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Toxicity associated with organic pollutants in aquatic sediments has not been fully characterized for the major estuarine and marine systems of coastal Louisiana. As such, five inshore and three offshore transects of the Mississippi River delta were sampled for sediment concentrations of polycyclic aromatic hydrocarbons (PAHs) and environmental variables. From 2012 to 2014, five environmental variables were measured in the northern Gulf of Mexico including: sediment organic carbon, dissolved oxygen and temperature of the overlying water column, and the percent sediment moisture and percent organic matter of the aquatic sediment. A two-step, response-variant regression model (GLM) was performed to determine significant effects of measured environmental parameters. The most important environmental variables affecting the concentrations of the measured compounds were those describing the characteristics of the aquatic sediments. The percent moisture of the sediment was the most important environmental parameter, significantly affecting eight of the ten organic pollutants and all three toxicity indicators. Percent organic matter of the sediments was the second most significant parameter, accounting for the variability in concentration for five of the measured pollutants. Temperature was significant for three of the PAHs and TEQ, and in every instance it had a negative effect on concentration. Dissolved oxygen of the water column was a significant variable on the concentration a single organic compound. Water column depth and salinity did not have a significant effect on the concentration on any of the constituents. The most significant environmental variables accounting for the variability of sediment PAH concentrations, included sediment moisture and organic matter. The physiochemical properties of the overlying water column had little to no significant effect on the concentrations of the majority of the measured pollutants. This research suggests that sediment organic parameters and their measurements at each sampling location should be incorporated into monitoring study design in order to more completely interpret the sediment burden of organic pollutants in aquatic sediments.

**WE326** Microbiological resistance to chemical pollution by urban effluents might be triggered by desiccation events.

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Freshwater ecosystems are subjected to different anthropogenic pressures. Among them, wastewater treatment plant (WWTP) effluents can represent a significant proportion of total discharge, mostly in regions suffering from water scarcity. WWTP effluents contain a mixture of assimilable and toxic compounds, which concentration ultimately determines the effect of the mixture on freshwater biota. Moreover, overexploitation of water resources together with climate change-driven pressures is causing drought events leading to desiccation to increase in number and intensity. To date, little is known about how desiccation events shape the response of river microbial communities to WWTP effluents. The present study used 24 experimental channels in a replicated regresional design to evaluate how an acute desiccation event shapes the response of a complex microbial community (i.e. a river biofilm) exposed to a dilution of a WWTP effluent. We found that desiccation and wastewater effluent significantly affected bacterial community and key biofilm processes such as photosynthesis, denitrification and methanogenesis. After the desiccation event, the biofilm associated to coarse sediment showed a stimulatory effect even at low dilution factors, which was not observed under control (i.e. no desiccation) conditions. Our results seem to indicate a simplification of the biofilm community after the desiccation event and a subsequent co-tolerance phenomenon. We argue that the acute desiccation event...
reduced diversity, selecting for resistant species. These resistant species could benefit from low dilution factors of wastewater effluent. Given that microbial metabolism powers biogeochemical cycling in ecosystems, we argue that functioning of freshwater ecosystems may be shaped as a result of the combined action of climate change-related stresses such as desiccation and chemical pollution.

WE327 Synergy effects of fluoxetine and variability in temperature lead to proportionally greater fitness costs: A multigenerational test

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Increased variability in water temperature is predicted to impose disproportionately greater fitness costs than mean increase in temperature. Additionally, water contaminants are currently a major source of human induced stress likely to produce fitness costs. Global change models forecast an increase in these two human induced stressors. Yet, in spite the growing interest in understanding how organisms respond to global change, the joint fitness effects of water pollution and increased variability in temperature remain unclear. Here, using a multigenerational design, we test the hypothesis that exposure to high concentrations of fluoxetine, a human medicine commonly found in freshwater systems, causes greater lifetime fitness costs when associated with increase variability in temperature. Although flight fitness and survival for both single and multiple stressors increased. Interestingly, fluoxetine and variability in temperature had no effect on the probability of survival. Freshwater systems are of the most imperiled ecosystems, often being confined to multiple human induced stressors. Our results indicate that organisms face greater fitness risk when exposed to multiple stressors at the same time than when each stress acts on alone. We observe the importance of using a multi-generational approach to fully understand individual environmental tolerance and its responses to a global change scenario in aquatic systems.

WE328 Influence of extreme heat events in the recovery capability of Mytilus galloprovincialis exposed to mercury contamination

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Several studies already described the impacts caused by metals in estuarine species, including mussels, but very scarce information is available regarding their effects in a global warming context. It is, however, unclear how temperature changes can also affect the response of organisms but their capacity to recover from pollution events. In this way, the present study aimed to understand the impact of warming in the capacity of Mytilus galloprovincialis to recover their biochemical performance after being pre-exposed to Hg. For this, mussels were exposed during 14 days at 17°C in the absence or presence of Hg (17°C; 17°C Hg), after which mussels were exposed during 7 days at 21°C in the absence or presence of Hg (O2: 31.2°C; 21°C Hg) or kept for the same period at 17°C in the absence or presence of Hg (17°C; 17°C Hg), and biomarkers related to mussels’ metabolic and oxidative stress status were evaluated as well as Hg bioconcentration. Our findings revealed that independently on the temperature regime, organisms previously exposed to Hg followed by a 28 days period in the absence of Hg were able to significantly decrease their metal concentration. Furthermore, energy-related and oxidative stress markers in mussels exposed for 28 days in the absence of Hg demonstrated no differences between mussels exposed to warming conditions (21°C) and control temperature (17°C), with a tendency to reach control values (observed in mussels exposed the entire experiment to 17°C in the absence of Hg).

WE329 Impacts of ocean warming and BDE-209 contamination on the energy budget of juvenile white seabream (Diplodus sargus)

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Climate change and chemical contamination are global environmental threats of growing concern for the scientific community and regulatory authorities. Yet, the impacts and interactions of both stressors (particularly ocean warming and emerging chemical contaminants) on the physiological responses of marine organisms remain unclear and require further research. In this context, our main goal was to study, for the first time, the effects of warming (+5°C, i.e. 24°C) and accumulation of a polybrominated diphenyl ether congener (BDE-209, brominated flame retardant) by dietary exposure on the energy budget of juvenile white seabream (Diplodus sargus; 3.5±2 g total body), used as a model. Specifically, growth (G), routine metabolism (R), excretion (faeces, F) and nitrogenous losses, U and food consumption (C) were calculated to obtain the energy budget. The results demonstrated that the energy proportion spent for growth dominated the mode of the energy allocation of juvenile white seabream (50.0-67.8%), including even under the synergistic effect of warming and BDE-209 exposure. On the other hand, energy loss via faeces was significantly higher under control temperature and BDE-209 exposure (16.0%). In all treatments, the energy channelled for metabolism was around 26% and a smaller percentage was channelled for excretion (faeces: 4.3-16.0% and ammonia: 2.3-3.3%). In general, the parameters were significantly affected by increased temperature and exposure to the flame retardant, with higher levels found under warming conditions (for wet weight, relative growth rate, protein and ash contents), BDE-209 exposure (only for O:N ratio) or under both stressors (for ammonia excretion rate and routine metabolic rate). On the other hand, lower viscoseromax index (VSI) was observed under warming and lower fat content was observed under both stressors. Overall, with such extreme and expected conditions of warming and contamination, the energy budget of marine fish species is expected to be greatly affected, leading to impacts on fish fitness and ecosystem functioning. Hence, the joint action of warming and chemical contamination could be considered as a powerful mechanism to enhance species resilience to projected warming. However, it is unknown how exposure to pesticide under warming in the parental generation will shape the offspring susceptibility to these stressors, specifically in vector species. We studied the transgenerational effects of single and combined exposure to warming (4°C increase) and the pesticide chlorpyrifos on life history traits and antipredator behaviors of the vector mosquito Culex pipiens using a bifactorial transgenerational experiment. Parental exposure to either warming or the pesticide, had negative effects on the offspring: both parental exposure to warming and to the pesticide resulted in an overall lower offspring survival. Parental warming impaired the anti-predator behaviors of the offspring by decreasing the diving proportion and diving time off the offspring. Within both the parental and the offspring generations, warming made the pesticide more toxic in terms of survival. However, this synergism disappeared in the offspring of parents who had been exposed to both stressors simultaneously because in this condition the pesticide was already more lethal at the lower temperature. For anti-predator behaviors, in both generations the two stressors reduced diving time in a synergistic way. In the parental generation, the effect of pesticide were stronger at 20°C than at 24°C. In the offspring generation, this synergetic effect depended on parental treatment. Pesticide induced stronger reduction in diving time at 20°C than at 24°C but only in the offspring of parents exposed to 20°C. Our results indicate that transgenerational effects will not increase the ability of this vector species to deal with pesticides in a warming world. This study highlights the importance of using bifactorial transgenerational experiment to understand the combined impact of pesticide and warming across generations, hence to assess the efficacy of vector control in a warming world.

WE330 Transgenerational effects of pesticide on vector mosquito Culex pipiens under global warming

T. Tran, L. Janssens, K.U.Leuven; K.V. Dinh, DTU Technical University of Denmark; R. Stoks, University of Leuven / Department of Biology

Recent transgenerational studies have showed that some species could acclimate to warming and pesticide separately. Transgenerational plasticity is even being considered as a powerful mechanism to enhance species resilience to projected warming. However, it is unknown how exposure to pesticide under warming in the parental generation will shape the offspring susceptibility to these stressors, specifically in vector species. We studied the transgenerational effects of single and combined exposure to warming (4°C increase) and the pesticide chlorpyrifos on life history traits and antipredator behaviors of the vector mosquito Culex pipiens using a bifactorial transgenerational experiment. Parental exposure to either warming or the pesticide, had negative effects on the offspring: both parental exposure to warming and to the pesticide resulted in an overall lower offspring survival. Parental warming impaired the anti-predator behaviors of the offspring by decreasing the diving proportion and diving time off the offspring. Within both the parental and the offspring generations, warming made the pesticide more toxic in terms of survival. However, this synergism disappeared in the offspring of parents who had been exposed to both stressors simultaneously because in this condition the pesticide was already more lethal at the lower temperature. For anti-predator behaviors, in both generations the two stressors reduced diving time in a synergistic way. In the parental generation, the effect of pesticide were stronger at 20°C than at 24°C. In the offspring generation, this synergetic effect depended on parental treatment. Pesticide induced stronger reduction in diving time at 20°C than at 24°C but only in the offspring of parents exposed to 20°C. Our results indicate that transgenerational effects will not increase the ability of this vector species to deal with pesticides in a warming world. This study highlights the importance of using bifactorial transgenerational experiment to understand the combined impact of pesticide and warming across generations, hence to assess the efficacy of vector control in a warming world.
2-parametric non-linear mixed effect model was used to describe nauplii development over time (**Instar = K / (1 + (K - 1) * exp (- (exp (log(mu) + age)))))**, where **K** is the asymptotic development stage and **mu** is the average stage transition rate). Effects of treatment and pedigree on the two model parameters were examined by comparison of models with likelihood ratio tests and Akaike’s Information Criterion (**AIC**). This analysis finds that treatment influenced developmental stage at the end of the experiment, while pedigree affected the time to reach it. Developmental effects were found in an increment of S in development stages of surviving individuals. When all individuals in control had reached maturity (288 hours), individuals exposed to the combined treatment kaikorine + copper were significantly delayed compared to all other treatments. Effects on individuals in the combined treatment were greater than expected based on the two treatments alone. A positive effect on development was already evident at the time of the first emerging copepodes (138 hours). These results indicate a synergistic relationship between risk of predation and copper by increased age at maturity in developing individuals of *T. brevicornis*. The results also show the significant role of pedigree in determining development rate. This highlights the need to consider both natural stressors and individual heterogeneity when conducting ecotoxicological studies.

**WE332 Functional and structural soil-vegetation indicators of ecosystem functioning in metal-contaminated environments: a case study in SE Spain**

J. Alvarez-Ragel, A. Pechalver Alcalá, M. Tercero Gómez, H. Conejo Alcaraz, O. Muthrue Ogque, Estela Telchir M. Universidad Politécnica de Cartagena / Ciencia y Tecnología Agraria; F. Jiménez-Cáceres, BIOCYMA, Consulting in Environment and Quality, S.L. Murcia, Spain.; C. van Gestel, Vrije Universiteit Amsterdam / Ecological Science; M. González-Alcaraz, Departamento de Biology & CESAM - University of Aveiro / Biology & CESAM Functional and structural soil parameters of six environments defined by stages of vegetation development were studied. The range of radiation doses included a disconnection of other lines of evidence (e.g. ecological modelling results) to predict long-term effects and toxicity. Some aquatic insect taxa (Chironomini, Chironomidae) were found to be highly sensitive to neonicotinoid concentrations under Mediterranean conditions. The lowest calculated NOECs from this study are below 0.2 µg/L for imidacloprid and for the neonicotinoid mixture, indicating that the current water quality criteria proposed by regulatory agencies and recent scientific publications (0.2 µg/L) may be underprotective for lentic aquatic ecosystems under Mediterranean conditions. However, due to the high variability of the results at the low test concentrations, the hypothesis must be confirmed by performing more specific tests.

**WE334 Multiple stressor effects of ionising (γ) radiation and non-ionising (UV) radiation on the IR duckweed (Lemma minor)**

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In nature environment, aquatic biota is facing to the ionizing radiation emitted from nuclear industry and radioactive waste. The effects may also be enhanced by radiation from human activity such as nuclear power plants accident, nuclear medicine and weapon tests. (Van Hoek et al. 2015). Among different ionizing radiation types, the toxicity of high dose gamma (γ) radiation is frequently studied in different aquatic organisms such as mammals, fish, crustaceans, higher plants, and algae. However, there is still lack of knowledge about the toxic effects of low dose γ radiation. The irradiation experiments in the study conducted in this work was to study the combined effect of low-dose γ-radiation (13.2, 20.3 and 47.1 mGy/h) and UVR (UVB 0.5 mW/m²) in the aquatic plant duckweed (*Lemma minor*) using a combination of genomic, functional and adverse toxicity endpoints. The results indicate that single γ-radiation reduced *L. minor* reproductive rate at a high dose (47.1 mGy/h, 7.9 Gy) after 7 days’ exposure. At the cellular level, γ-radiation inhibited photosystem II (PS II) maximal efficiency (Fv/Fm) and oxidative phosphorylation (OXPHOS) and enhanced the non-photochemical quenching (NPQ), light-saturated PS II operating efficiency (Fv/Fm), electron transport rate (ETR) and reactive oxygen species (ROS) formation. Single UVR caused similar effects as IR and additionally induced morphological change (size and colony disconnection) in the plant. When exposed in combination, enhanced reproductive inhibition, OXPHOS reduction, PSII inhibition, NPQ and ROS formation were observed for the high γ-radiation dose (47.1 mGy/h). Antagonistic effects on Fv/Fm, pigments content, photochemical quenching (qP) and ROS formation were observed at low to intermediate γ-radiation doses (13.2 and 20.3 mGy/h). Meanwhile, the simultaneous exposure of IR and UVR enhanced the toxicity of IR and UVR. The results are consistent with the hypothesis that radiations and chemicals are currently on going to identify the most relevant toxic pathways being perturbed by the single and multiple stressors tested. Combined study with radiations and chemicals are currently on going.

**WE335 Natural organic matter determines the potential of titanium dioxide nanoparticles to mitigate pesticide toxicity in presence of UV light**

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Nano-particle-based technology has evolved to a global industry with a tremendous economic potential. Since 2006, the investment in nanotechnology increased from estimated $1.18 billion to more than $2.5 trillion in 2015. Among all manufactured nanoparticles (NPs), titanium dioxide NPs (nTiO₂) belong to the most frequently used and produced NPs. As a consequence of their incremental use, nTiO₂ will end up in surface waters. The aim of the present study was to evaluate if light triggers the photocatalytic potential of nTiO₂ to form reactive oxygen species (ROS). ROS have the ability to reduce the toxicity of chemical stressors in aquatic invertebrates. The role of ubiquitous natural organic matter (NOM) for this process is still unclear. Therefore, the objectives of this work were to study the combined effect of low to intermediate γ-radiation (13.2 and 20.3 mGy/h) and NOM on the toxicity of several pesticides included in the mixture as compared to imidacloprid. Therefore, it may be concluded that the CA model provides an accurate prediction for short-term effects at the population and community levels but requires the inclusion of other lines of evidence (e.g. ecological modelling results) to predict long-term effects and toxicity. Several aquatic insect taxa (Chironomini, Chironomidae) were found to be highly sensitive to neonicotinoid concentrations under Mediterranean conditions. The lowest calculated NOECs from this study are below 0.2 µg/L for imidacloprid and for the neonicotinoid mixture, indicating that the current water quality criteria proposed by regulatory agencies and recent scientific publications (0.2 µg/L) may be underprotective for lentic aquatic ecosystems under Mediterranean conditions. However, due to the high variability of the results at the low test concentrations, the hypothesis must be confirmed by performing more specific tests.
toxicity of Primicarb was reduced 1.7-fold with increasing UV (0.00 vs. 2.20-2.60 W/m²). In presence of NOM, Primicarb toxicity was generally decreased (up to 2.3-fold, e.g. 0.00 W UV/m²). Depending on pesticide type and factor combinations we observed both positive and negative effects of UV radiation on the toxicity of the selected pesticides. A general prediction on the combined effects of nTiO₂, NOM, and UV on the toxicity of pesticides seems currently difficult. Rather, physicochemical properties like pesticide structure, solubility, adsorption, and desorption seem to play a major role in the interaction with nTiO₂, NOM, and UV-A, and the ultimate pesticide toxicity.

WE336
Effects of inorganic sunscreen formulations on the algal symbionts of reef building corals, Symbiodinium spp., and their combined toxicity with ocean warming
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Between 4,000 and 6,000 tons of sunscreens annually are washed from the skin by swimming and diving, and directly released into the oceans, posing a potential threat to coral reef ecosystems. Titanium dioxide nanoparticles (nTiO₂) are common UV filters in cosmetic products, and in typical sunscreen formulations they are dispersed in an oil phase, a mixture of emulsifiers and emollients which can reach up to 20% of the product content by weight. In order to assess the impact of inorganic sunscreen on corals’ endosymbiotic algae (Symbiodinium spp.), three nTiO₂ with different sizes and surface coatings were dispersed in an oil phase to mimic commercial available sunscreen formulations. Two Symbiodinium phylotypes, known for their different tolerance to environmental change and stress, were exposed to oil:nTiO₂ dispersions at both ambient temperature (26°C) and thermal stress condition (32°C). Growth rates, maximum photosynthetic activity and reactive oxygen species (ROS) production were evaluated. Symbiodinium spp. exposure to sunscreen show negative effects on all endpoints studied and the toxicity is enhanced with simultaneous heat stress. Results indicate toxicity is not dependent on the type of nanoparticle and it is likely driven by the oil carrier, a major ingredient in all cosmetic sunscreens. Released oil, and the nanoparticles in the oil droplets, accumulate in the water surface microlayer and sediments due to its lipophilic characteristics and resistance against particle removal, constituting a major risk to marine organisms. Corals rely on the photosynthate provided by the endosymbiotic algae for their nutrition, and the production of excess ROS by Symbiodinium cells as a consequence of heat stress, is considered to be a trigger of coral bleaching (the loss of Symbiodinium from the coral host). The significant decrease of maximum photosynthetic activity at 32°C coupled with the algae growth depression and direct release of ROS into the water, poses a potential threat to coral reef ecosystems. The increased ROS production following sunscreen exposure, in addition to the reduction of photosynthetic activity, provide evidence that exposure to these types of sunscreens may exacerbate bleaching response in corals and pose a risk to coral reef ecosystems in a changing ocean.

WE337
Metallothioneins as an indicator of metal exposure in a naturally mineral enriched aquatic environment
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The Marico River, in the North West province of South Africa, is relatively unaffected by anthropogenic activities. However, metal concentrations – mainly from natural sources – occasionally exceed environmental quality guidelines or toxic concentrations. Macroinvertebrates are capable to react to these metals through processes such as the induction of metallothioneins (MTs). The aims of this study were to determine whether the induction of MTs can be used as indicator of natural metal exposure in anthropogenically impacted systems, and whether there are relationships between metal concentrations in water, sediment and macroinvertebrates and concomitant MT levels. This was done by sampling macroinvertebrates, water and sediment from eight sites in the Marico River and tributaries. Water and sediment samples were prepared and analysed with an ICP-MS to determine metal concentrations. Seven families including Notonectidae, Coenagrionidae, Atyidae, Libellulidae, Baetidae, Caenidae and Chironomidae were selected and digested for metal analyses. Four families including Gyrinidae, Aeshnidae, Atyidae and Coenagrionidae were also analysed to determine their MT concentration. To test for possible relationships between metal concentrations in the macroinvertebrate families, MTs and metal concentrations in the water and sediment, Spearman’s non-parametric correlation tests were done among sites. Positive correlations were found between metals in sediment and macroinvertebrates, while there were no correlations between metal concentrations in water and macroinvertebrates. Even in freshwater river systems with a relatively low human impact and no mining activities, a positive correlation existed between trace metal bioaccumulation (e.g. Ni, Pb, Zn) in macroinvertebrates and the induction of MTs. There were, however, no correlations between MTs and bioaccumulation of earth metals (e.g. Al, Fe, Mn, Ti). These data clearly demonstrate the application value of MTs as markers for metal exposure in freshwater ecosystems.

WE338
Mollusks as indicators of environmental pollution (case studies in marine mussel Mytilus galloprovincialis Lam. and terrestrial snail Bradybaena fruticum Mull.)
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Snails are widely used as sentinel species in marine and terrestrial biomonitoring. Their wide distribution in different biotopes, high sensitivity to pollution makes them good bioindicators in environmental quality assessment. The comparability of mollusks in pure and polluted sites was performed in field studies round Moscow Region and in Moscow city as well as for marine bioindication in a few locations round Sevastopol city. Cardiac activity registration in selected mollusks was carried out as a tool for measuring deterioration of general health of local biota in the sites of the concern. Methodology with standard testing of heart rate (HR) and HR variability using functional loads (short-term temperature, salinity change, some mechanical stimuli, etc.) were carried out. In land biodynamics a 3-4 years old genetically modified morphs of bush snails Bradybaena fruticum (Mull.) were used collected from Moscow city (Kartmazoov, Kuz/minky,Ismailov Park) with respectively HR of one site and compared with in Moscow city (Kartmazoov, Kuz/minky) differed in low thermosteresis from those of the reference side and Ismailovskoye Park demonstrated in dynamics of HRs. The second part of the study is 2007-2008 a few populations of benthic molluscs (bush snails, Pila), was performed in Sevastopol Bays using salinity change test. Possible links between chemical pollution by dioxin (in terrestrial snails) and heavy metals contents in mollusk’s tissues (in marine mussels) and peculiarities of HR recovery after thermal or salinity loads were discussed.

WE339
The effect of temperature on toxicity of cypermethrin on Daphnia magna
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Cypermethrin is an insecticide which European Parliament of the Council has classified as a priority substance in Directive 2013/39/EU in the field of Water Policy. The Annex II of the Water Policy sets the environmental quality standards (EQS) for the priority substances. Cypermethrin’s annual average and maximum allowable concentration in inland surface waters are 0.06 ng/L and 0.6 ng/L respectively, and annual average and maximum allowable concentrations in environmental quality standard. The abiotic stressor like the temperature can have an effect on toxicity of the chemical. Previous studies have shown that the temperature of the environment affect the toxicity of pesticides belonging to the pyrethroids which cypermethrin is part of. The 48-hour half maximal concentration (EC50) and median effective time (ET50) values were tested with custracean Daphnia magna immobilization at the temperatures 10°C, 16°C and 20°C in laboratory experiments. Cypermethrin was almost twice as toxic at 10°C (2.17 ± 0.20 µg/L) compared with 20°C (4.10 ± 0.30 µg/L). The EC50 value of 16°C was 2.64 ± 0.21 µg/L which was closer to EC50 value of 10°C than 20°C. The temperature had statistically significant effect in the EC50 experiment. The temperature did have a slight effect in the ET50 experiment. The ET50 value at 10°C was 30.60 ± 0.74 hours compared with 33.12 ± 0.79 and 32.86 ± 0.83 hours respectively at 16°C and 20°C. The only statistically significant difference between the temperatures was between 10°C and 16°C. The temperature dependent behavior of cypermethrin was not taken into account when the environmental quality standard for cypermethrin was set by European Union and the United States Environmental protection agency’s reregistration eligibility decision for cypermethrin. With pyrethroids deltamethrin and permethrin the Canadian guidelines do not take account effects of temperature. When guidelines and legislations are prepared to certain chemicals some abiotic factors like the temperature should be considered. Key to changing guidelines and legislation is to take account the possible effects of temperature in test standards.

WE340
Pattern oriented food web modelling of metal mesocosm datasets
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The risk assessment of metals has a long history and over time a large collection of
ecotoxicity data has been accumulated. The most informative tests performed for the ecological effects of toxicants are mesocosm studies: controlled experiments where the effects of toxicants on model communities are studied for extended periods of time. Mesocosm studies are cost- and labor-intensive but offer a unique insight into realistic ecological effects of toxicants: they address not only direct effects on sensitive species, but also indirect effects resulting from ecological interactions (e.g., competition, predation) between sensitive and less sensitive species. Typically, the effects occurring in mesocosms are more complex and difficult to interpret. A study has been set up to investigate whether food web modelling can reveal additional patterns, trends, or interactions in existing copper and zinc mesocosm datasets. Pattern oriented food web modelling – an ecological modelling technique – is used to reveal the mechanisms underlying metal effects. When analysing multiple patterns in the datasets will be identified and described. Models are then evaluated for their ability to reproduce these patterns. In the case of mesocosm studies, food web models can be applied to understand the mechanism behind observed patterns. A large diversity of food web models exists with large differences in their complexity and underlying theories. Food web models based on ordinary differential equations are relatively simple in structure while they can still account for the interactions between species in the food web. They are therefore ideally suited to study mesocosm data. Additionally, the effects of environmental parameters such as temperature and pH, often key determinants of metal toxicity, can be included. Finally, the potential Pathway of Effects – explaining how metal toxicity leads to the observed patterns – can be evaluated to identify the most important drivers of metal toxicity in the mesocosm food webs and derive community-based threshold levels.

**WE341** Bioaccumulation and physiological conditions in *Ruditapes philippinarum* from the Vallona lagoon (northern Adriatic Sea, NE Italy): Application of Contaminant/shell weight indices

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*Ruditapes philippinarum* (Adams & Reeve, 1850)is a soft-bottom dweller bivalve commonly used in biomonitoring programs, especially in bioaccumulation assessment, owing to its high tolerance to toxic compounds. However, bioavailability and accumulation of contaminants in the soft tissue of molluscs could be affected by abiotic factors, such as food availability, pH and temperature, and also by biotic factors, such as the seasonal changes of flesh weight in molluscs.

In this context, some issues could arise especially when comparing different sites in a short-term biomonitoring with data obtained from different periods of the year. In this study, bioaccumulation of metals, Polyyclic Aromatic Hydrocarbons (PAHs) and Butyltins (BTs) in Manila clams was monitored twice a year, from November 2010 to June 2015, in order to assess impact of human activities on *R. philippinarum* population from the Vallona Lagoon, a transitional area located in the delta of the Po River (NE, ITALY) which is the largest and most important Italian watercourse and one of the main sources of contaminants to Adriatic environments. Although levels were quite consistent with those reported from other geographical areas with low to medium pollution, seasonal trends were showed for each contaminant with higher concentrations on autumn rather than on spring surveys. The physiological condition of clams was also examined through two indices (condition index and survival in air) and they both exhibited seasonal variations in correlation with the physicochemical parameters of water (temperature, pH, salinity and dissolved oxygen) were also considered. To ensure that bioavailability assessment was not affected by seasonal variation of soft tissues of molluscs, Contaminant/shell weight indices, which consist of normalization achieved multiplying each contaminant concentrations by the condition index, were applied. Accordingly, the normalization enabled us to highlight the contaminant uptake from clams in some particular periods and to compare different sites in a long-term biomonitoring with data obtained from different periods of the year. Indeed, some contaminants showed quite a steady state all over the monitoring period and at the different sites, whilst others, such as Arsenic, Chrome, Nickel, Lead, Copper, Zinc and BTs, showed different patterns of bioaccumulation with some periods presenting enhanced concentrations probably related to anthropogenic activities.

**WE342** Biomonitoring of Singapore mangroves using biomarker expression and contaminant burden in caged green mussels, *Perna viridis*. G. Juhel, National University of Singapore; S. Bayen, McGill University / Singapore-Delft Water Alliance; E. Segovia, C. Goh, W. Lee, National University of Singapore; B.C. Kelly, National University of Singapore / Civil & Environmental Engineering

Mangroves are fragile coastal ecosystems whose ecological and socioeconomic importance for adjacent ecosystems and local populations is being clearly recognized nowadays. Mangroves are one of the most threatened tropical environments and our understanding of the impact of chemical pollution on these ecosystems is still at its infancy. In this study, nine mangrove sites were selected around Singapore coastline to cover various contamination profiles, and green mussels, *Perna viridis* were used as bioindicators. Bivalves were deployed in cages at each of the nine mangrove patches for 28 days on two consecutive years (one year during the monsoon period and the other year during a dry-weather period), and collected for subsequent analysis. A series of biochemical and cellular biomarkers were developed and measured using various organs. Metallothioneins (MTs), Glutathione-S-Transferase (GST), Ethoxyresorufin O-deethylase activity (EROD), Vitellogenin-like protein (VLP), and Activity of Acetylhymesterase (AChE) measured in the bivalves’ haemocoel tissues. Mussel’s haemolymph was also used to evaluate immunological parameters (Total Haemocyte counts, phagocytosis and lysozyme levels) and the level of haemocytos’ DNA damage, using the Comet assay. Results of this study revealed different profiles of biomarker expression between the various sites. Most notably, metallothionein induction was observed at some the sites, indicating potential exposure to heavy metals while higher levels of DNA damage and EROD were also recorded at some of the mangrove patches pointing towards possible exposure to organic contaminants. Some biomarkers appeared to be subject to seasonal variations while others were very stable. Possible correlation between biomarker expression and the level of various contaminants (metals, PAHs, pharmaceuticals, endocrine disruptors chemicals, personal care products) in caged mussels were also studied. Using an Integrated Biomarker response index, the various mangrove sites were eventually ranked amongst each other. Our findings ultimately indicated a clear segregation of mangrove sites, indicating that some mangrove patches were potentially more at risk than others towards chemical contamination.

**WE343** Impacts of climate change on mercury bioaccumulation in large ocean predators

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Many marine ecosystems are being affected by climate driven changes in freshwater discharge, circulation, productivity and seawater temperature. Large ocean predators such as tunas account for a large proportion of methylmercury exposure in many countries and almost 40% of U.S. population wide intake between 2010-2012). Methylmercury is a potent neurotoxin, particularly for children, and has been associated with impaired cardiovascular health in adults. We have developed a new mechanistic model for methylmercury bioaccumulation in marine food webs (BAM). The model is driven by methylmercury concentrations in seawater and ocean biogeochemical conditions (temperature, dissolved organic carbon, DOC, and trophic status). We apply this model to investigate the magnitude of changes in methylmercury concentrations expected from decadal oscillations in seawater temperature in the North Atlantic. We compare the magnitude of these changes to those occurring in the Pacific Ocean and discuss how climate related variability is likely to affects exposures of humans and wildlife to methylmercury. Our results suggest changes in tissue burdens driven by oscillations in seawater temperature are similar in magnitude to those that have been achieved by reductions in emissions in the North Atlantic.

**WE344** Chemical stress on aquatic communities under semi-arid conditions: towards an improved multimetric approach

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Combined effects of chemical pollution and water scarcity are one of the most important threats to aquatic ecosystems in (semi-)arid regions of the world. The low dilution potential of chemicals (urban, industrial and agricultural) discharged in aquatic ecosystems under water scarcity conditions could lead to devastating toxic effects. Moreover, a large proportion of aquatic bodies in these regions are known to have a high hydrological variability in a temporal scale. In this sense, communities naturally adapted to this condition are expected to respond differently to toxic stress than those adapted to more constant water flows. Here we apply this model to investigate the magnitude of changes in methylmercury concentrations expected from decadal oscillations in seawater temperature in the North Atlantic. We compare the magnitude of these changes to those occurring in the Pacific Ocean and discuss how climate related variability is likely to affects exposures of humans and wildlife to methylmercury. Our results suggest changes in tissue burdens driven by oscillations in seawater temperature are similar in magnitude to those that have been achieved by reductions in emissions in the North Atlantic.
a temporal and spatial scale were evaluated at a taxonomic and at a biological trait level. Significant responses to multiple stressors from some invertebrate taxa and functional traits (feeding habits, reproduction and respiration) were determined. Based on these results, suggestions for a biological vulnerability multivariate index, which considers more site-specific conditions, will be presented.

WE345 Long-term effects on transplanted caged-freshwater bivalves Diplodon chilensis to the assessment of water quality in a Patagonian river
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Population growth and human activity are contributing to habitat deterioration in Patagonian rivers with the consequent threat to water quality and biodiversity. The bivalve Diplodon chilensis is a key species in the macroinvertebrate fauna of Patagonian lakes and rivers which has been proposed as a sentinel organism in the area. We evaluated the long-term effects of transplantation of caged D. chilensis to different sites in the Chimehuin river (reference site (S1), downstream from an aquaculture facility (S2), and downstream from an open dump and from the sewage treatment plant (S3)) after 3, 6, 9 and 12 months of exposure. We combined the antioxidant response, oxidative damage, ROS production and energetic status, with water and sediment analysis (physico-chemical and biological variables, and organic matter content). Physico-chemical variables varied according to site and time of exposure. Sites S2 and S3 showed generally higher chlorophyll a concentration and total coliform bacteria values compared to site S1, whereas organic matter content in the sediment was elevated only at site S2. In D. chilensis, gill SOD and GST activity was higher in both S2 and S3 than in S1 by the end of the exposure time. During the last month of exposure (month 12), GSH levels dropped dramatically and lipid peroxidation levels increased in individuals from S2 and S3 sites. Digestive gland factor (DFG) and energy values in digestive gland were increased at sites S2 and S3, from 6 to 9 months of exposure. Our results indicate that despite the large flow rate of Chimehuin river water quality is deteriorated in areas impacted by anthropogenic activities. This effect is reflected by a physiological response of D. chilensis, which is especially significant during period of their highest metabolic activity (autumn fall/ winter).

WE346 The influence of selected seasonal and anthropogenic phenomena on a perennial river in South Africa.
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The quality of surface waters worldwide is declining fast. This is due to anthropogenic stressors such as a change in temperature and an increase in toxicants and nutrients, which are predominant abiotic agents of disturbance in intermittent streams. The objective of this study was to establish whether seasonal fluctuations can reduce the effect of anthropogenic impacts on the river and whether this is reflected by macroinvertebrate assemblages, physico-chemical water parameters and metal concentrations in the water and sediment. Water quality and macroinvertebrate community data were collected in the Crocodile River (South Africa). Four surveys were conducted, two during the low-flow and two during the high-flow season. Spatial and temporal variations were found with regard to suspended solids and total organic material. The low flow surveys had a slight increase in suspended solids and total organic material, whereas a substantial increase occurred during the high-flow season. The highest concentrations of metals in the water column were in contrast to the sediment concentrations observed during the high-flow seasons at the majority of the sites. Only minor differences in sediment metal concentrations were noted between high and low flow seasons. The pH values increased from the origin of the river downstream. However this did not result in a clear trend with regard to either an increase in sediment metals or a decrease in the concentration of dissolved metals. A combination of biological indices (Taxa Richness, Shannon-Wiener diversity index and Pielou’s evenness index) were calculated for each site and season, and no significant differences were found between the high and low flow seasons for any of the indices at each of the sites (P>0.05). The highest percentage of families at all the sites and seasons were classified as highly tolerant and tolerant to organic enrichment. It can be concluded that the high flow seasons (associated with rain and floods) did not have a rejuvenating effect on the river, as mentioned in previous studies. This phenomenon is substantiated by the metals concentrations, total organic matter, total suspended solids, electrical conductivity and the fact that all the taxon collected in large numbers during both seasons were tolerant to highly tolerant.

WE347 Growth, Photosynthetic and Antioxidative Defense System Response of Hordeum vulgare to Combined Stress of Heat Wave and Drought
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Extreme climatic events such as heat waves and drought periods are predicted to increase in frequency and severity in many regions under future climate scenarios, and in natural environment these two abiotic stresses often occur simultaneously. The short-term (3 day-long) impact of +10 °C heat wave treatment 6.5 h per day was investigated on Hordeum vulgare under well-watered and water deficit conditions in Closed-top chambers under controlled environment. The decreases in shoots dry weight, shoots length and leaves area were observed in the water deficit treatment after exposure to heat wave, while all these parameters in the well-watered treatment were not affected significantly. The decline in growth under water deficit conditions was most likely caused by a considerably greater reduction in photosynthetic rate as well as far stronger oxidative stress caused by combined impact of heat wave and drought than from single heat wave treatment as revealed by higher level of malondialdehyde content and considerably stronger stimulation of antioxidative enzymes. Full recovery of physiological processes and water content in plants occurred 1 day after heat wave treatment after one day-regeneration period. In contrast, neither shoots dry weight nor leaves area as well as most physiological processes analyzed, membrane damage, and catalase activity in water deficit treatment were not recovered to the control value. The obtained results showed that drought-stressed Hordeum vulgare plants suffered markedly stronger physiological and oxidative stress caused by combined stressors as compared to the respective controls and revealed an important of soil water availability even during the short-term heat wave period. Keywords: Hordeum vulgare, heat wave, drought stress, antioxidant system, photosynthesis, growth

WE348 Does elevated CO2 protects plants against heat waves damage?
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The frequency and severity of heat waves is increasing as a result of climate change. The extreme events may lead to decreased crop productivity and financial incomes. Differential responses of crops and weeds to heat waves and CO2 may also cause shifts in their competitive interactions. The aim of the study was to examine the influence of extreme events (heat wave plus drought) and CO2 on the growth of barley Hordeum vulgare L. and wild mustard Sinapis arvensis L. Barley and wild mustard, growing together in the microcosms at the combination 2:1, were subjected to short-term processes and were compared with controls (2°C under controlled environment). The obtained results showed that drought-stressed Hordeum vulgare plants suffered markedly stronger physiological and oxidative stress caused by combined stressors as compared to the respective controls and revealed an important of soil water availability even during the short-term heat wave period. Keywords: Hordeum vulgare, heat wave, drought stress, antioxidant system, photosynthesis, growth

WE349 Combined effects of increasing temperatures, drought and an insecticide on freshwater zooplankton communities: a microcosm study
A.A. Sanchez, IMDEA Water Institute / Aquatic Ecotoxicology; I. López, L. Nozal, IMDEA Water Institute; M. Vighi, IMDEA Water Institute / Earth and Environmental Sciences; A. Rico, IMDEA Water Institute / Aquatic Ecotoxicology Water scarcity and chemical pollution are two of the main groups of stressors causing ecological impairment in freshwater aquatic ecosystems of (semi-)arid regions. Predictions about human and climatic pressure on water resources in these regions reveal that the interaction between these two groups of stressors will increase in the nearby future. In line with that, advances in ecological risk assessment recognize that stress factors harming aquatic ecosystems rarely operate individually. Therefore, new approaches to assess interactions between multiple stressors are needed. In this study we evaluated the combined effects of the insecticide lufenuron and two additional stress factors: increasing water temperature and drought. Twenty-seven microcosms were stocked with pond water, sediment, and a homogeneous plankton assemblage. Three environmental scenarios were simulated: 20°C and 28°C without desiccation, and 28°C with desiccation. The experiment was performed in triplicate with three insecticide dosages (Control, Low, High Concentration) in each environmental scenario. The insecticide was applied twice, with a 10 day interval between applications. Test units without desiccation were refilled twice per week with distilled water. Test units exposed to drought stress were not refilled and allowed to desiccate. After that, these units were refilled up to the initial level and maintained for two more weeks. Lufenuron concentrations in water and sediment were periodically measured, together with water quality parameters (DO, pH, T, EC and nutrients). Zooplankton composition was determined on a weekly basis, and the isolated and interactive effects of temperature, drought and lufenuron were evaluated using suitable statistical techniques. Effects were assessed at the community and at the
population level. Lufenuron was the main stressor in all the environmental scenarios, with a significant decrease of Cladocera and Copepoda, and an increase of Rotifera. Temperature and drought had slight effects on community composition and accelerated insecticide dissipation, influencing community recovery capacity. Interaction between factors at community and population level was mainly observed at the beginning of the experiment. Direct and indirect responses at population level varied between environmental scenarios. The results of this study contribute to understand differences in vulnerability of aquatic ecosystems to multiple stressors in (semi-)arid regions.

**WE350** Toxicity of phenoxy herbicide: the effects of elevated temperature and CO2 concentration

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Climate change is a major concern for agriculture and crop productivity. Crop productivity strongly depends on crop protection measures such as use of herbicides. Climate change will influence the fate and ecotoxicity of herbicides by altering their environmental partitioning and degradation, distribution and abundance of weeds and growth and development of weeds and crops. Differential responses of crops and weeds to elevated temperature and CO2 may also cause shifts in the co-evolution of interactions. The aim of the study was to to examine the influence of elevated temperature and CO2 on the effects of phenoxy herbicide to spring barley (Hordeum vulgare L.) and common lambsquarters (Chenopodium album L.). Two climate scenarios were investigated: current climate (21 °C, 400 ppm CO2) and future climate (25 °C, 800 ppm CO2). The terrestrial target Ch. album and non-target H. vulgare plants, growing together in the microcosms at the combination 2, were sprayed with herbicide sprays solutions equivalent to 0.5-2 of field application rate. The plants were sprayed at the four- to five-leaf stage with herbicide solutions prepared with 4-Chloro-2-methylphenoxyacetic acid (MCPA, CAS No. 94-74-6, Sigma-Aldrich). The growth and response of antioxidant defence system of both species were evaluated.

**WE351** Combined effects of insecticide exposure and predation risk on freshwater detritivores

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Exposure to sub-lethal concentrations of insecticides are known to pose at risk non-target insects due to effects on physiology and behaviour. Under natural conditions, a food web is usually in a dynamic equilibrium with a diversity of biological interactions and natural stressors, such as predation. Perception of predation risk alone may change the behaviour and physiology of prey species, impacting their fitness and, thus, alter their susceptibility to chemical exposure. There are thus growing efforts to understand how the combined effects of toxicants and biotic stressors may affect populations, their web dynamics and ecosystem functioning. Chlorantraniliprole (C. riparius) and diuron (C. boltonii) were chosen as a model compound an anthranilic diamide largely applied due to its specificity for insect ryanodine receptors of target species. So, to assess the combined effects of insecticide exposure under predation risk on freshwater detritivores we studied the behavioural and developmental responses of Chironomus riparius. Plus, we tested whether the responses of the C. riparius, a collector, would change in the presence of a predator species and the associated production of fine particulate organic matter (FPOM). For that, trials were performed using a simplified trophic chain: Alnus glutinosa L. as a model species and the collector Chironomus riparius and its natural predator the dragonfly Cordulegaster boltonii. A full factorial design tested the effects of the CAP (0 or 2 µg/L), presence/absence of the predator C. boltonii and of the herbicide diuron (0.5 mg/L and 1 mg/L) on the FPOM loss and FPOM production. These results showed that the combined herbicide stressors impair some physiological aspects of the predator. C. riparius growth rate was thus decreased independently by all factors (CAP exposure, predation risk and predator presence) and a marginal interaction between CAP and predation risk was also observed. To conclude, this study highlights the need to consider natural biotic stressors and species interactions in risk assessment of chemical pollution, since both vertical and horizontal diversity play their role on response to stress.

**WE352** How sugarcane and high temperatures are contributing to amphibian declines in Brazil? Morphological, biochemical and molecular approaches

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Sugarcane is the most efficient first-generation source of ethanol in the market, which has contributed to the rapid expansion of its crops and generated concerns related to its environmental impacts to Brazilian territory. In tropical areas, agricultural activities of sugarcane are intensified during the rainy season, which coincides with the period of occurrence of the amphibian species. Tadpoles and other aquatic animals from tropical areas of the world also experience large temperature fluctuations in their habitats. So, there is a great concern that amphibians are not only being affected by pesticides in their environments, but also by the combined effects of pesticides and temperature changes. In this study, we evaluated biochemical, morphological and molecular effects caused by the exposure to herbicides used on sugarcane crops in Brazil (diuron and its metabolite, 3,4-DCA, clomazone and sulfentrazone) on tadpoles of different species, using different thermal gradients. Our results showed that temperature is an important factor influencing the toxicity of pesticides in tadpoles. Diuron combined to higher temperatures accelerated metamorphosis process in tadpoles of Lithobates catesbeianus. Temperature and herbicide concentration (deo2, dio3, thibz, tra, tfb and klf9) were mostly upregulated in these groups, showing disrupting effects of diuron for amphibians. 3,4-DCA presented similar responses to diuron on L. catesbeianus and its effects were also pronounced at high temperatures. Native tadpoles of Rhinella schneideri and Euphylus nattereri had their antioxidant defense system affected by exposure to the herbicides clomazone and sulfentrazone (C. Boltonii and Thiamethoxam). Changes between species may also cause shifts in the co-evolution of interactions. The aim of the study was to examine the influence of elevated temperature and CO2 on the effects of phenoxy herbicide to sugarbarley (Hordeum vulgare L.) and common lambsquarters (Chenopodium album L.). Two climate scenarios were investigated: current climate (21 °C, 400 ppm CO2) and future climate (25 °C, 800 ppm CO2). The terrestrial target Ch. album and non-target H. vulgare plants, growing together in the microcosms at the combination 2, were sprayed with herbicide sprays solutions equivalent to 0.5-2 of field application rate. The plants were sprayed at the four- to five-leaf stage with herbicide solutions prepared with 4-Chloro-2-methylphenoxyacetic acid (MCPA, CAS No. 94-74-6, Sigma-Aldrich). The growth and response of antioxidant defence system of both species were evaluated.
pesticide ingredients. Temperature, rainfall, soil parameters, pest ranges and cropping patterns are all expected to alter under climate change conditions, and all of these parameters could affect pesticide use and environmental exposure patterns which will alter the risk that these compounds pose to the environment. Here, we report the results of a study to evaluate the impacts of climate change on the exposure of aquatic systems in the UK. Concentrations of a number of case study pesticides, covering a range of physico-chemical properties and uses, were modelled in streams and rivers in the UK using two models recommended by the Forum for the Co-ordination of Pesticide Fate Models and their Use, namely MACRO and PRZM. Model predictions were obtained for current conditions and for expected conditions in the 2080s by parameterising the models for current and future predicted weather conditions, cropping patterns, soil properties and pesticide use patterns. The results show that exposure patterns of the APIs are likely to change in the future. These changes in exposure vary by pesticide type and the region modelled with exposure in some chemical-region combinations increasing and in others decreasing.

WE355
Ranking micropollutants in effluent by exposure indices evaluated via suspect/nontarget screening
P. Narke, Changwon National University / Environmental Engineering; c. younghun, Changwon National University / FEED of Eco-Friendly Offshore Structure; J. Jeon, Changwon National University / Environmental Engineering Information on the occurrence and concentration of micropollutants in effluents from wastewater treatment plants (WWTPs) provides important clues for evaluating the risk and availability of river ecosystems. However, the chemical monitoring work for the numerous trace contaminants is time-consuming, labor-intensive, and cost a lot. To overcome the problems, the efficient monitoring programs have been conducted for limited, but prioritized pollutants. In general, the prioritization has mainly been based on effect/toxicity information rather than exposure. Relatively, pollutants with high occurrence frequency and concentration have often been underestimated when their effect/toxicity are neither considerable nor well defined. In the present study, a list of prior effluent micropollutants is suggested with a exposure-index based scoring/ranking procedure following qualitative chemical analysis. A scoring table with exposure indices such as occurrence frequency and chronology peak area was applied for the ranking. WWTP effluent samples were taken in September, 2016 and analyzed via suspect/nontarget screening using LC-HRMS (QExactive+ Orbitrap). Within a suspect list, about 60 compounds were tentatively identified and ranked by the score. After purchasing reference standards for high rankers, about 20 micropollutants were orthogonally confirmed and roughly quantified. The prioritized micropollutants were mostly pharmaceuticals and personal care products including 9 groups such as analgesics/anti-inflammatoryatories (acetaminophen, mefenamic acid), antibiotics/antifungal (climbazole, fluconazole, sulfamethoxazole, sulfamethazine), anticonvulsant (carbamazepine, carbamazepine-epoxide, oxcarbazepine), antihistamines (diphenhydramine, fexofenadine), antihypertensive agent (ibresetan, valsartan), antiasthmatic (albuterol), and lipolytics (acetadoline). All the concentrations for the top ranker, acetaminophen detected in all 7 samples, was ranged up to 1,300 ng L⁻¹. The 2nd ranking pollutant was caffeine and followed by cimetidine≥ mefenamic acid≥fexofenadine>carbamazepine>ibresetan>fluconazole≥depethphalhydrine≥ sulfamethoxazole. Since some tentatively identified pollutants were left unconfirmed, the prioritized compound list should be updated along with additional confirmations. Nevertheless the ranked list still include highly exposable micropollutants which are worthy for intensive monitoring in effluents.

WE356
Interspecific effects of temperature shifts on life parameters, oxidative stress, and expression of fatty acid synthesis genes and heat shock protein genes in two congeneric copepods Tigriopus sp.
J. Hua, Sungkyunkwan University / Biological Science; J. Lee, Sungkyunkwan University
In this study, we compared the effects of temperature changes on lifecycle parameters, intracellular reactive oxygen species (ROS), ATP content, glutathione S-transferase (GST) enzyme activity, and gene expression profiles of both the de novo lipogenesis (DNL) pathway and heat shock protein (hsp) genes in the temperate copepod Tigriopus japonicus and the Antarctic copepod Tigriopus kingsejongensis. The median lethal temperature (LT50) and no observed effect level (NOEL) in T. japonicus were determined to be 35.3°C and 32°C, respectively, in 48 h for temperature of 2°C per day. In T. kingsejongensis, LT50 and NOEL were determined to be 24.8°C and 12°C, respectively. Levels of OS and GST activity were slightly elevated (<e>P

WE357
Effects of water bathing on zooplankton physiology and fitness driven by food characteristics in a long-term enclosure experiments
L. Minguez, LIEC (CNRS UMR 7360, Université de Lorraine) / Laboratoire Interdisciplinaire des Environnements Continentaux, CNRS UMR 7360; E. Sperfeld, University of Oslo / Centre for Ecological and Evolutionary Synthesis (CEES), Department of Biosciences; S.A. Berger, J.C. Nejstgaard, Leibniz Institute of Freshwater Ecology and Inland Fisheries / Dept. Experimental Limnology; M.O. Gessner, Leibniz Institute of Freshwater Ecology and Inland Fisheries
Ecotoxicological assays using Daphnia species are generally performed under optimal food and light conditions. However, results of such assays may not adequately reflect stress responses in the wild, since the ability of organisms to cope with adverse conditions critically depends on the amount of available energy. One type of potential stressors is increasing warming in recent years, which is a terrestrial-derived dissolved organic carbon (tDOC) that causes browning of lakes and streams, but long-term effects of tDOC on freshwater organisms are not sufficiently known. Using a combination of an in situ enclosure experiment and laboratory incubations, we tested whether long-term tDOC exposure affects the physiology and life-history traits of the water flea Daphnia longispina, and whether any observed effects are reversible. Daphnids were collected from a long-term, large-scale enclosure experiment conducted in a clear-water lake in northeastern Germany, where a natural plankton community was exposed to a standard source of tDOC (HuminFeed). The physiological state of daphnids in the enclosures was followed after addition of the tDOC. In the first week of exposure, daphnids experienced oxidative stress, i.e. an imbalance in favour of oxidative damage, but this response was no longer observed after 36 days. Daphnids and water from the enclosures sampled again after 10 weeks were used to assess survival and reproductive performance under laboratory conditions. Both survival and reproduction were related to sex traits and the elemental and biochemical composition of the daphnids. Surprisingly, daphnids kept in brown water showed higher survival and fertility than their counterparts kept in clear water (A). This unexpected outcome is explained by higher sex ratio and quality in enclosures receiving tDOC, related to a higher abundance and biomass of nutrients food algae. Moreover, transplantation of daphnids from A to B enclosure water and vice versa revealed considerable plasticity, as the daphnids were capable of rapidly adjusting their metabolism to a similar level as that observed in the species tests that only allow for the assessment of direct pesticide impacts. Thus, our study points to the importance of accounting for exposure duration and food quality and quantity when assessing Daphnia responses to environmental stressors such as tDOC.

WE358
Interactive effects of multiple stressors on estuarine processes
A. O’Brien, The University of Melbourne; K. Dafforn, Macquarie University / Evolution and Ecology Research Centre; M. Mayer-Pinto, University of New South Wales / Evolution Ecology Research Centre School of Biological Earth and Environmental Sciences; E.L. Johnston, University of New South Wales / Evolution and Ecology Research Centre; A. Charron, Macquarie University / Molecular Ecology and Toxicology
Natural systems are threatened by a variety of different anthropogenic stressors. These are often considered in isolation, but in reality most are found in combination and have the potential to interact with different outcomes. Urban systems such as estuaries and harbours are commonly exposed to chemical (e.g. contaminants), physical (e.g. built infrastructures) and biological (e.g. invasive species) stressors. It is important to understand the interactive threats posed by these stressors. Here we use a systematic literature review to explore selected urban stressors and their potential interactions under current environmental conditions. We link the interactive effects to responses in key ecological processes including biogeochemical cycling, primary production and ecological interactions. Using functional endpoints that are common across systems we have the ability to identify similar patterns and trends. This allows us to go beyond comparisons that rely on community structural endpoints and are potentially only relevant at local scales.

WE359
Ecology or reproducibility crisis? - Lessons from a laboratory scale tri-trophic test system
V. Riedl, Environment Department, University of York / Environment Department; A. Agatz, IBACON GmbH / Environment Department; R. Benstead, Fera Science Ltd / Centre for Chemical Safety and Stewardship; R. Ashauer, University of York / Environment
Concerns of reproducibility in toxicology are becoming increasingly important. Such concerns have been raised regarding a lack of reproducibility in scientific research. There is indeed evidence that a number of research findings are not reproducible by others or even within the original laboratories. Yet, while the reproducibility of results might often be difficult, it is essential in the context of regulatory decision making. In the environmental risk assessment of pesticides, for instance, replication, standardization and reproducibility are of great importance to the final decision. The level of statistical power and the robustness of test findings. For this reason, rapid single-species tests that only allow for the assessment of direct pesticide impacts are still more frequently used than multi-species systems. Although they are ecologically more relevant, micro-/mesocosms often yield lower statistical power due to higher complexity, difficulty of standardization, resource demand and variability among replicates. However, growing evidence suggests that direct effects measured at the individual level do not proportionally translate to effects at the population and community level. The use of testing procedures that are ecologically more realistic but ideally comply with regulatory needs should thus be a priority to risk assessors and regulators. The tri-trophic aquatic test system
TriCosm (P. subcapitata, Ceriodaphnia, Hydra) was developed as an intermediate link between single-species tests and complex multi-species systems, to detect small stressor-induced alterations in ecological interactions. The achievement of standardization, replication and reproducibility was given close attention during the development of the system, yet, the TriCosm was found to be compliant in terms of repeatability and reproducibility only in the short term. Here we present experiments designed to discern effects of variation due to ecologically influencing factors that impact on the community dynamics in this aquatic multi-species system.

WE360 Improving tolerance to natural and chemical stressors by inducing early life stages in the rotifer Brachionus sp. Cayman

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Rotifers are widely used as bio-indicators and models for ecotoxicology due to characteristics such as high ingestion rate, rapid growth, ease of culture in small volumes, ease of establishing clone cultures, short generation time, small size, and sensitivity to various toxicants. The monogonont rotifer Brachionus plicatilis (Müller, 1786) is a euryhaline species, typically found in salt lakes and coastal brackish waters, presenting high commercial importance and value, since it is commonly used as live feed for several marine species larvae in aquaculture productions. In the last decade it has become commonly accepted that environmental stimuli can induce phenotypic alterations in the organisms. Thereby, the primary objective of this project was to assess if temperature shocks during early life stages of rotifer development could improve their tolerance to different natural and man-made stressors. Temperature was used both as acute toxicity bioassays and 48h chronic toxicity bioassays were conducted with two strains of Brachionus sp. Cayman (MRS10 and BA3), a biotype within the B. plicatilis complex, obtained from the Laboratory of Aquaculture and Artemia Reference Center (Ghent University, Belgium). The bioassays were performed in accordance with the International Standards ISO 19820 and ISO 20666, respectively, using 25°C and 25°C as control environmental conditions. The organisms were exposed to different concentrations of antibiotics, pesticides, and oxidative stress inducers, as well as to different salinity and temperature conditions. Consequently, to test for possible increased tolerance to these stressors, neonates (0-4h) were exposed to short non-lethal temperature shocks (cold and heat), and after a recovery period, survival tests were performed. The sensitivity values compared. Results show that temperature shocks during early life stages result in changes in the sensitivity of rotifers to different stressors. These are very promising results, and their application in an aquaculture context could be advantageous to decrease the mass mortality in rotifer production. Future research within this project will address possible epigenetic mechanisms (DNA methylation or histone modifications) behind those genetic changes and assess if their maintenance can be achieved through several generations.

WE361 Effects of a mixture of pharmaceuticals in a freshwater model ecosystem

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Owing to their ecological importance, freshwaterers provide important services which leads to a strong societal demand concerning the preservation of their ecosystem. They are the receptors of many contaminants emitted by human activities and more specifically, Polywaste water treatment plants effluents. Water resources of the Meuse watershed are used on both sides of the French-Belgian border which involves a coherent and joint management. In that framework, the DIADEM project suggests developing and spreading out a cross-border multidisciplinary approach to improve the diagnosis and the chemical and biological (biomarkers) monitoring of freshwaterers using the Meuse river as a case study. In the past, results from chemical surface water monitoring of the Meuse has revealed the occurrence of numerous substances and more particularly pharmaceuticals. Overall, the project suggests coupling chemical and biomarkers analysis on caged organisms (a crustacean, a mollusk, a moss and a fish species) with predictive mathematical population level distributions (SSDs) for aquatic organisms also provide valuable additional information for higher-tier testing strategies for risk assessments for plant protection products. While typically only the standard end-point (e.g. 96-hour LC₅₀) is used, the thorough analysis of existing studies (i.e. individual tests or combined knowledge from different studies) as well as possible adaptations of standard test designs at the organism group of concern may provide valuable facts like time-dependency or reciprocity of effects. In this perspective, the development of quantitative structure toxicity relationships (QSARs) between chemical hydrophobicity and toxicity have been shown for nearly 100 years in both mammals and fish, typically using the log of the octanol-water partition coefficient (kow). The current study reassessed the influence of mode of action (MOA) on aquatic toxicity-log kow relationships using a comprehensive database of curated and standardized acute toxicity and consensus log Kow values, and weight of evidence methodology. Log kow classifications were developed as linear regression of log acute toxicity and log kow for 50 different combinations of taxa (e.g., fish, invertebrates, species-specific) and MOA (6 broad; 3 specific narcosis subtypes). MOA categories included narcosis (non-polar, polar, ester), acetylene-lactonerase inhibition, neurotoxicity, electron transport inhibition, ionosomeregulatory/circulatory impairment, and reactivity. Forty-eight of the 50 MOA-based models were statistically significant (p < 0.05; most p < 0.001), but r² values were generally less than 0.5, particularly for non-narcosis MOAs. The results showed that MOA-based QSAR models can improve the accuracy of aquatic toxicity predictions for a range of taxa, and that incorrect classification of a specific acting chemical can result in toxicity prediction errors greater than 1000 fold.

WE364 Data-mining: Making use of aquatic lower-tier data for higher-tier risk evaluation of agrochemicals

G. Eck, U. Memmert, E. Eschenbach, Eurosifins Regulatory AG.

Apart from delivering relevant toxicity data, standard lower-tier toxicity studies on aquatic organisms also provide valuable additional information for higher-tier testing strategies for risk assessments for plant protection products. While typically only the standard end-point (e.g. 96-hour LC₅₀) is used, the thorough analysis of existing studies (i.e. individual tests or combined knowledge from different studies) as well as possible adaptations of standard test designs at the organism group of concern may provide valuable facts like time-dependency or reciprocity of effects. In this perspective, the development of quantitative structure toxicity relationships (QSARs) between chemical hydrophobicity and toxicity have been shown for nearly 100 years in both mammals and fish, typically using the log of the octanol-water partition coefficient (kow). The current study reassessed the influence of mode of action (MOA) on aquatic toxicity-log kow relationships using a comprehensive database of curated and standardized acute toxicity and consensus log Kow values, and weight of evidence methodology. Log kow classifications were developed as linear regression of log acute toxicity and log kow for 50 different combinations of taxa (e.g., fish, invertebrates, species-specific) and MOA (6 broad; 3 specific narcosis subtypes). MOA categories included narcosis (non-polar, polar, ester), acetylene-lactonerase inhibition, neurotoxicity, electron transport inhibition, ionosomeregulatory/circulatory impairment, and reactivity. Forty-eight of the 50 MOA-based models were statistically significant (p < 0.05; most p < 0.001), but r² values were generally less than 0.5, particularly for non-narcosis MOAs. The results showed that MOA-based QSAR models can improve the accuracy of aquatic toxicity predictions for a range of taxa, and that incorrect classification of a specific acting chemical can result in toxicity prediction errors greater than 1000 fold.

WE365 Comparison of models and tools for derivation of species sensitivity distributions (SSDs) for use in pesticide risk assessment

L. Azevedo, BASF SE, Agrarzentrum Limburgerhof / Global Toxicology; G. Eck, U. Memmert, E. Eschenbach, Eurosifins Regulatory AG.

Apart from delivering relevant toxicity data, standard lower-tier toxicity studies on aquatic organisms also provide valuable additional information for higher-tier testing strategies for risk assessments for plant protection products. While typically only the standard end-point (e.g. 96-hour LC₅₀) is used, the thorough analysis of existing studies (i.e. individual tests or combined knowledge from different studies) as well as possible adaptations of standard test designs at the organism group of concern may provide valuable facts like time-dependency or reciprocity of effects. In this perspective, the development of quantitative structure toxicity relationships (QSARs) between chemical hydrophobicity and toxicity have been shown for nearly 100 years in both mammals and fish, typically using the log of the octanol-water partition coefficient (kow). The current study reassessed the influence of mode of action (MOA) on aquatic toxicity-log kow relationships using a comprehensive database of curated and standardized acute toxicity and consensus log Kow values, and weight of evidence methodology. Log kow classifications were developed as linear regression of log acute toxicity and log kow for 50 different combinations of taxa (e.g., fish, invertebrates, species-specific) and MOA (6 broad; 3 specific narcosis subtypes). MOA categories included narcosis (non-polar, polar, ester), acetylene-lactonerase inhibition, neurotoxicity, electron transport inhibition, ionosomeregulatory/circulatory impairment, and reactivity. Forty-eight of the 50 MOA-based models were statistically significant (p < 0.05; most p < 0.001), but r² values were generally less than 0.5, particularly for non-narcosis MOAs. The results showed that MOA-based QSAR models can improve the accuracy of aquatic toxicity predictions for a range of taxa, and that incorrect classification of a specific acting chemical can result in toxicity prediction errors greater than 1000 fold.
WE366 Effects on NTa communities: HCx vs NOEC design
F. M. Bakker, Eurofins-Mitox; S. Aldershof, Bioresearch and Evaluation
We discuss two examples of field fauna study designs with non-target arthropods (NTa). In both cases a hay meadow was chosen as a paradigm representative for off-field habitats at risk. One example concerns an HC, approach where EC, for various x were estimated from a field experiment and used to derive a Species Sensitivity Distribution. The other example concerns a more “classical” approach where a limited number of species were tested in a replicated block design and the NOEC endpoint was found via statistical hypothesis testing. The merits and caveats of both approaches will be discussed and a protocol for evaluating and documenting statistical and biological significance of a NOEC study design will be presented.
We analyse whether “No Effects” may have statistical or biological causes. In the HCx-study consistent dose-response curves were obtained within 4 major arthropod taxa with 128% of the 63 curves 776; 80%) and the results could be constructed for each of them. Due to full overlap of curves the classes could be combined, resulting in narrow confidence intervals. In the NOEC-study 69 from 596 (11%) taxa were valid for univariate analysis, representing all major arthropod taxa. The protocol developed for the classification of results yielded an informative evaluation and allowed results to be classified as inconclusive or conclusive on a confidence scale of 1-4. Both study designs were fit for purpose and Type I and Type II errors that lead to low effect dose levels, respectively. This contribution challenges the dominance of α and underscores the prominence of β when it comes to consumer safety. After all, a false positive result in a regulatory context implies a potential economic loss (re-testing, lost market share, lack of appropriate protection agents), whereas a false negative result implies a risk to environmental health. Statistical insignificance (P>F > α) does not imply biologically and statistically valid results. Where the HCx-design was statistically straightforward, the regulatory implication of the findings was not. On the other hand, where the regulatory implication of the NOEC is clear, the methodological issues related to hypothesis testing hamper a clear-cut presentation. An evaluation summary table, involving MSĐ-analysis, as presented here may be helpful in this regard.

WE367 α-Dominance versus β-Prominence
F. M. Bakker, Eurofins-Mitox; S. Aldershof, Bioresearch and Evaluation
The NOEC or an equivalent regulatory set ECx values are key endpoints to assess safety and to protect consumers challenges & tested in a replicated block design and the assessment takes place in a multispecies context, e.g. in non-target arthropod (NTa) field studies. To date most ecotoxicological faunistic NTa field studies follow a hypothesis test design. Few examples can be found that address ECx-finding by extrapolation from a regression model, such as the SSD-curve. There are two risks associated with hypothesis tests: the producers’ risk and the consumers’ risk, better known as Type I and Type II errors that lead to low effect dose levels, respectively. This contribution challenges the dominance of α and underscores the prominence of β when it comes to consumer safety. After all, a false positive result in a regulatory context implies a potential economic loss (re-testing, lost market share, lack of appropriate protection agents), whereas a false negative result implies a risk to environmental health. Statistical insignificance (P>F > α) does not imply biologically and statistically valid results. Where the HCx-design was statistically straightforward, the regulatory implication of the findings was not. On the other hand, where the regulatory implication of the NOEC is clear, the methodological issues related to hypothesis testing hamper a clear-cut presentation. An evaluation summary table, involving MSĐ-analysis, as presented here may be helpful in this regard.

WE368 Defining simple toxicity values (EC, BMD) is not so simple
E. Bizaret, Université de Lorraine, CNRS UMR 7360; F. Latras, Helmholtz Center for Environmental Research - UFZ GmbH; V. BAILLARD, LIEC (CNRS UMR 7360, Université de Lorraine); S. Devin, LIEC, CNRS UMR 7360, Université de Lorraine / LIEC, CNRS; M. Schmitt-Jansen, UFZ - Helmholtz Centre Environm. Research / Department of Biocatalytic Ecotoxicology; M. Delignette-Muller, VetAgro Sup / Laboratory of Biometry and Evolutionnary Biology
Effective Concentrations (ECs) have now largely supplanted No Observed Effect Concentrations (NOEC), after decades of statistical criticisms towards the latter. ECx has a simple definition which sounds unambiguous. However, depending on the concentration-response pattern, its derivation is not trivial and should be paid attention in the context of ecotoxicological risk assessment. We recently developed a workflow for high-throughput concentration-response modelling of omics data (e.g. transcriptomics, metabolomics). Such data often displayed non-monotonic trends (U or Umbrella shape) as well as linear and exponential trends. Based on our results, sigmoidal concentration-response shape was more than the exception than the rule, as also reported in the literature for such omics data. In this context, we will discuss and explain why these non-sigmoidal trends lead to several issues regarding the derivation of toxicity values. In particular, the derivation of EC does make sense only provided an asymptotic response level is observed at high exposure concentrations and, in the case of non-monotonic trends, requires the definition of a maximal amplitude of the response. Alternatively to EC, the Benchmark Dose Concentration (BDx) has been proposed in the context of ecotoxicology for setting toxicity values. The BMD approach as mentioned in EFSA guidance proposes two options. The first one considers a x-fold change of the control response which seems hazardous sensitive to the signal level (if the control response is zero, so will be the x-fold change). The second option defines a critical response level accounting for the standard deviation of data (control response + SD). We will illustrate why this feature makes it more robust and useful whatever the concentration-response model.

WE369 Calculating the true ECx/LCx for non-linear models
Z. Gao, Bayer AG Crop Science Division; A. Solga, Bayer AG; H. Frenndt, Bayer AG Crop Science Division; T. Preuss, Bayer AG / Environmental Safety
ECx and LC50 are most frequently used endpoints for deriving Predicted No-Effect Concentration (PNEC) or Regulatory Accepted Concentration (RAC). ECx is defined as the concentration that shows x% effect compared to the control. This concentration is at which 50% mortality was observed. These definitions inherently assume the adverse effect at control is 0%. For example, in terms of mortality, the mortality at control should be 0% and when there is background mortality, Abbot’s correction has to be applied. In terms of plant growth, percentage of inhibition in growth compared to control is calculated to feed in the dose-response model. These approaches have bypassed the requirements to use the standardly used probit dose-response model by modifying the data to make the model assumptions valid. However, use of these approaches without caution can cause serious over- or under-estimation of EC/LCx due to the ignorance of control variability, the improper use of binomial assumption behind the probit model, etc.
On the other hand, it is nowadays recommended that nonlinear regression models shall be used for dose-response analysis for metric data, which creates another problem in practice because there is inconsistency in the definition of ECx in regulatory context and that in the software implementing these 3 or 4 parameter models procedures. The ECx in programming implementation often assumes the 100% effect is the difference between the baseline response at control and the maximum response at dose level of infinity. EC50 is then defined as the 50% effect between baseline and maximum effect calculated from the model, not with respect to control as how it is defined. This can lead to misinterpretations in the context of regulatory risk assessment. In this study, simulation examples and real data examples will be presented to illustrate the impact of the misuse of the current standard dose-response analysis procedures.

WE370 Review of Dose-Response Analyses in Regulatory Framework
Z. Gao, Bayer AG Crop Science Division; A. Solga, Bayer AG; H. Frenndt, Bayer AG Crop Science Division; T. Preuss, Bayer AG / Environmental Safety
Low effect value ECs (EC10, EC20) derived from dose-response models have been recommended to replace NOECs in the pesticide regulatory context (e.g., new Regulation (EC) No. 1107/2009). The use of NOEC is often criticized because the standard dose power to detect NOECs and the derivation of null hypothesis testing can be very low due to high variability and small sample size. However, the concept and the limitations behind the various dose-response models have not been systematically addressed. There are ambiguities in the terminologies used such as linear and nonlinear dose-response models. When to use which model is not clear to practitioners. Practical difficulties in the implementation of the methodology lead to questions like what to do when there are no monotonic dose-response relationships, when ECs is superior to NOEC and when NOEC is more appropriate, why the confidence intervals are very broad in the range of low effect dose levels, and so on. In this study, we provide an in-depth review of the various dose-response models and associated assumptions and indications to answer these questions. Challenges in which a certain dose response model is more appropriate than others were described and illustrated using both real and simulated data examples. We show that the type of data, quantal, count or continuous are important to determine the error structure in the statistical model and the data characteristics provide inherently hints in the choice of dose-response model. The shared parametersizations and curve shapes between the so-called linear and non-linear models are clear in the model structure and are emphasized here. We also identify a few common mistakes in practice due to wrong interpretation of dose-response analysis or wrong understanding of the software implementations. Potential improvements after the decision tree approaches proposed in the EFSA Guidance are discussed. The knowledge gaps related to non-monotonic dose-response relationships are also tackled. The connection between the multiple comparison procedure to derive NOEC and the model-based dose-response analysis are presented and hybrid approaches are discussed.
Deriving no effect levels using probabilistic approaches: Application to trichloroethylene (TCE) and potential impacts to risk-based exposure concentrations

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Derived no effect levels (DNELs) are indispensable tools needed to quantitatively evaluate the impact of various chemical exposures to humans and inform decisions related to exposure mitigation and environmental remediation. Typically, DNELs are calculated using deterministic methods that rely on single point estimates of no-effect levels, assessment factors (AFs) that allow extrapolation to human exposure scenarios and account for uncertainties in toxicological information, and allowable risk level. However, the point estimates used to calculate DNELs are by definition conservative estimates that when combined lead to a phenomenon termed “compounded conservatism”. The consequence of this phenomenon is DNELs that likely overestimate potential risks associated with human exposure to chemicals. Probabilistic risk assessment (PRA) approaches can be used to characterize the level of conservatism in deterministically-derived DNELs and to directly calculate DNELs. The advantage of calculating DNELs using PRA approaches is the ability to incorporate all available data and information associated with a particular input variable (i.e., variability amongst toxicity values, distributional assumptions of AFs, etc.) instead of relying on a single value, as is necessary for deterministic methods. An added benefit of the PRA approach is increased transparency regarding the protectiveness of a chemical’s DNEL. This work will demonstrate how PRA is used to calculate DNELs using trichloroethylene (TCE) as an example. The presentation will focus specifically on TCE’s non-carcinogenic effects and incorporate the variability and uncertainties associated with dose-response modeling, physiologically-based pharmacokinetic modeling, assignment of AFs, and the choice of allowable risk level. The potential impacts of using PRA approaches to calculate DNELs will be discussed in relation to resulting risk-based exposure concentrations.

WE372 Aquatic higher-tier exposure testing of pesticides - from complexity to simplicity

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Field exposure of pesticides is usually characterized by time-variable substance entries into water bodies resulting in complex exposure patterns which often significantly deviate from the constant exposure in standard ecotoxicity tests with aquatic organisms. As an appropriate risk refinement option higher-tier exposure testing is proposed in the current EFSA guidance document for aquatic risk assessments providing the possibility to define ecotoxicologically relevant test conditions and meaningful link to FOCUS exposure modelling outputs. Studies designed to reflect realistic exposure often result in lower effects. However, FOCUS exposure patterns are frequently challenged in regard to their representativeness for the variety of possible field scenarios and hence are generally not accepted as valid refinement option by several EU Member States. Besides, it is often difficult to generalize various critical patterns of different FOCUS scenarios for an efficient and to align these generic patterns with sensitive life stages of the organisms of concern. Practical aspects, types of effects and life traits of test organisms add to the complicity of designing reasonable higher-tier exposure studies. The challenge is to balance representativeness and practicability of test designs. Representativeness can for example be enhanced by multi-year modelling simulations where the variability of treatment and climate constellations are taken into account. Detailed analysis of the multitude of predicted exposure scenarios as well as a detailed analysis of available standard toxicity data is required to develop meaningful test designs and strategies. Generating simplicity from complexity rather than matching exactly the FOCUS modelling exposure pattern is proposed as potential solution to concerns on representativeness as well as practicability for ecotoxicity testing and finally acceptability in a regulatory context. The poster will present examples for the generation of reasonable test designs and strategies that are considered to meet objections towards higher-tier exposure testing as adequate means for refinement of aquatic risk.

WE373 Keeping it real: multidisciplinary approaches to aquatic risk assessment


Aquatic risk assessments for plant protection products (PPPs) can often be complex, comprising multiple crops, application rates, Member States (MSs) and therefore exposure scenarios. A critical to consider is that that hazard quotient (PEC/PNEC ratio) really represents and whether it is a realistic representation of the true risk. A number of assumptions and worst-case parameters are used within the Tier I risk assessment, both on the exposure (predicted exposure concentration; PEC) and the effect (predicted no effect concentration; PNEC) side of the equation. To maximise the realism within the risk assessment, it is therefore advantageous to take a multidisciplinary approach, involving specialists in environmental fate, exposure modelling, aquatic ecotoxicology testing and regulatory ecotoxicology when developing refinements for the risk assessment. By developing integrated solutions, it is possible to progress from theoretical to more realistic exposure scenarios and estimations of maximal concentrations that hazard quotients (PEC/PNEC ratios) really represents and whether it is a realistic representation of the true risk. A number of assumptions and worst-case parameters are used within the Tier I risk assessment, both on the exposure (predicted exposure concentration; PEC) and the effect (predicted no effect concentration; PNEC) side of the equation. To maximise the realism within the risk assessment, it is therefore advantageous to take a multidisciplinary approach, involving specialists in environmental fate, exposure modelling, aquatic ecotoxicology testing and regulatory ecotoxicology when developing refinements for the risk assessment. By developing integrated solutions, it is possible to progress from theoretical to more realistic exposure scenarios and estimations of maximal concentrations that hazard quotients (PEC/PNEC ratios) really represents and whether it is a realistic representation of the true risk.

WE374 Critical aspects of higher-tier laboratory exposure testing with different aquatic organisms

G. Gonsior, Eurofins Agrofination Services Ecotox GmbH; U. Memmert, G. Eck, E. Eschenbach, Eurofins Regulatory AG; C. Hafner, Eurofins Agrofination Services Ecotox GmbH / Aquatic Ecotoxicology

Revised exposure testing is an option for higher-tier risk evaluations proposed in the current EFSA guidance document for aquatic risk assessments for plant protection products. It offers scope for risk refinement by defining ecotoxicologically relevant concentrations that might be less conservative than constant exposure scenarios realised in standard effect studies or to justify time-weighted average concentrations in context with chronic risk assessments. Higher-tier exposure testing addresses the challenge of higher tier exposure scenarios as adequate to define representative exposures that are calculable using deterministic methods that rely on single point estimates of concentration; PEC) and the effect (predicted no effect concentration; PNEC) side of the equation. To maximise the realism within the risk assessment, it is therefore advantageous to take a multidisciplinary approach, involving specialists in environmental fate, exposure modelling, aquatic ecotoxicology testing and regulatory ecotoxicology when developing refinements for the risk assessment. By developing integrated solutions, it is possible to progress from theoretical to more realistic exposure scenarios and estimations of maximal concentrations that hazard quotients (PEC/PNEC ratios) really represents and whether it is a realistic representation of the true risk. A number of assumptions and worst-case parameters are used within the Tier I risk assessment, both on the exposure (predicted exposure concentration; PEC) and the effect (predicted no effect concentration; PNEC) side of the equation. To maximise the realism within the risk assessment, it is therefore advantageous to take a multidisciplinary approach, involving specialists in environmental fate, exposure modelling, aquatic ecotoxicology testing and regulatory ecotoxicology when developing refinements for the risk assessment. By developing integrated solutions, it is possible to progress from theoretical to more realistic exposure scenarios and estimations of maximal concentrations that hazard quotients (PEC/PNEC ratios) really represents and whether it is a realistic representation of the true risk.
Refined exposure tests have become part of the regulation framework for plant protection products in the EU (EFSA Aquatic Guidance Document 2013). A pulse dose test can be used to address areas of risk that cannot be satisfied with the standard suite of aquatic toxicity tests. A pulse dose considers situations where the expected exposure events in the field are significantly shorter than in the standard laboratory long-term tests. However, this challenge is often to cover all exposure scenarios from multiple scenarios within one test. Therefore, the maximum exposure (peak) concentration, the number of peaks, the duration of the peaks, and the interval between peaks are considered to simulate a realistic profile covering a large number of scenarios. In this study, three different life stages of rainbow trout (Oncorhynchus mykiss) were exposed to nine pulses of the test chemical. To set these pulses as sharp as possible, the fishes were transferred from treatment vessels to untreated vessels at each time of pulse application. All vessels, including controls, were kept under flow through conditions. The concentrations of the test chemical were measured at start and end of each pulse event. Fertilised eggs, newly hatched fry and juveniles, already swimming up, were exposed. Glass aquaria with a total volume of 30 L were used. The evaluation of biological effects was based on mean measured concentrations measured for the test substance pulses and could be compared with the predicted environmental concentrations based on FOCUS modeling simulations. In contrast to a continuous exposure, the procedure of several pulse applications may have an impact and possible impairment of the sensitive stages. However, it was demonstrated that the performance of the life stages exposed was acceptable and conforms to quality criteria set by the test guidelines (OECD, USEPA). The test design was shown to provide a suitable approach to address a very complex exposure regime to cover the ‘worst case’ when only a laboratory exposure is unrealistic.

**WE377**

**TIER2+: Developing the Tools for Future Risk Assessment - New Chronic Invertebrate Test Systems and the Application of Realistic Exposure Scenarios**

A. Dabrunz, F. Kümmerich, C. Lang, Eurofins Agroscience Services Ecotox GmbH / Aquatic Ecotoxicology

According to recent and proposed guidance of the European Food Safety Authority (EFSA) current aquatic toxicity test systems should be critically reviewed, adjusted in regards of realistic (e.g. pulsed) exposure and complemented by establishing new test systems. To account for these challenges we will present data on a twofold strategy including A) test systems and B) exposure conditions/ãn in Experimental data of newly established (sub)chronic test systems, which are suitable meeting Tier 2 regulatory requirements (EGLies), were used. The evaluation of biological effects was based on evaluated include, for example, ostracons, cycloids, nematodes, oigocheatas and amphipods, with a focus upon experimental conditions, suitability of standard toxicity testing endpoints and experimental variability. Subsequently, data on selected test systems under flow through test conditions simulating pulsed dose exposure scenarios will be given.

**WE378**

Optimisation of a chronic toxicity flow-through set up to investigate the adverse effects of chemicals to Daphnia magna

C. Beyer, IES Ltd; A. Peither, Innovative Environmental Services IES Ltd; S. Höger, Innovative Environmental Services (IES) Ltd / Environmental Toxicology; A. Krüger, University of Appl. Sc. Northwestern Switzerland / Institute of Ecopreneurship; P. Corvini, University of Applied Sciences Northwestern Switzerland

Daphnids are playing an important role as representative or indicator species for aquatic invertebrates in the environmental risk assessment for plant protection products, chemicals and pharmaceuticals. To investigate chronic toxicity, semi-static Daphnia magna reproduction tests following Test Guideline OECD 211 have to be performed. So far this test design is also used for highly degrading substances, despite the fact that by using semi-static test design the exposure concentration of the parent test compound is decreasing and metabolic products are accumulating during the renewal intervals of 2-3 days. To ensure a steady exposure level to avoid previously exposed of degradation products, the studies have to be performed in a flow-through system. We are presenting a new flow-through system for reproduction testing with Daphnia magna. The flow-through system may pose additional stress for the daphnids, since interval dosing of test medium is causing turbulences within the test vessels. To preclude that stress effects are given in the new flow-through system, a reproduction test with a hydrostatically stable test substrate was carried out. This comparative test allowed studying stress parameters by comparing the effect level concentrations and the ECn values. Furthermore, a reproduction test with a fast degrading substance was tested to prove the ability of the new flow-through system to maintain a parent compound concentration above 80% throughout the test. In the comparative test both test systems showed the same dose-response-curves and gave the same No Observed Effect Concentration and Lowest Observed Effect Concentration for continuous exposure per surviving test animals. Thus it can be assumed that the new flow-through system does not cause additional stress on Daphnia magna in a 21-day reproduction test. In the reproduction test with a non-degrading test item, the new flow-through system could dose the test item concentrations very reliable and precisely (dosed concentrations between 98 and 105% of nominal). In this study, it was proven that the new flow-through system does not cause unacceptable additional stress for the daphnids and can be utilized for reproduction testing with Daphnia magna. All tests conducted in the flow-through system were valid in accordance with OECD Guideline 211. The dosing system is very precise and reliable and is capable to maintain a parent compound concentration above 80% for a test item with half-life of 5 hours.

**WE379**

Eggs and larval fish test, an alternative method to marine fish exposure: Sensitivity and interest of early life stage.

F. Sanchez, C. Dupuy, A. Fournier, Groupe SGS France; J. Bertin, SGS Multisub / Ecotoxicology

Multi-trophic level bioassays are usually carried out to determine toxicity of effluents, chemicals, cosmetic ingredients, etc... Toxicity to species is different according to the compound type considered, therefore tests on plants, invertebrates and vertebrates should be conducted. On the other hand, vertebrate organism tests should be avoided when possible, including test on juvenile fish. Alternative methods need to be developed for juvenile fish testing, with equivalent predictability and sensitivity. Several alternative methods exist for freshwater studies but there is no standardized method available for seawater fish. This study is a first step in the evaluation of a marine fish embryo and larvae test as alternative to adult marine fish test OSPAR HOCNF guideline, for example. The embryo and larval sensitivit (toxicity of turbidity to differentiation and death of the test substance (larvae labras)) to reference substances were evaluated. To this purpose, within 72 hours post-fertilisation, the blastula stage eggs were exposed to reference substances separately. The effects of the toxicants on embryos and larvae were observed daily on a 10-days exposure period. For each reference substance and species, EC50 and mortality rate were calculated. To avoid vertebrate testing, results of this study were compared to published data. Several reference substances had a significant impact on survival of eggs and/or embryonic development. The sensitivity of the organisms is significantly different according to reference substances. Moreover, this test was used to evaluate toxicity of waste seawater samples in multi-trophic level bioassays (i.e. combination with single-species on algae, copepod and oyster larvae tests). Based on these results, marine fish embryo test appears as a credible alternative to juvenile fish testing. Therefore additional experiments will be conducted to validate this model.

**WE380**

Lack of Relevance of Normalized Hindlimb Length Measurement in Assessment of Thyroid Disruption in the Amphibian Metamorphosis Assay

S. Pawlowski, M. Dammann, S. Champ, BAFSE 5E; M. Mathis, P. Fort, Fort Environmental Labs, Inc.

The OECD Test No. 231 and USEPA Test Guidelines OPPTS 890.1100 (amphibian metamorphosis assay - AMA) represents a Tier 1 ecotoxicity test designed to evaluate thyroid disruption. The test exposes Nieuwkoop and Faber (NF) stage 41 Xenopus tropicalis to different amount and exposure of the test substance for 21-days and the following endpoints are measured: mortality, hind limb length (HLL), body length (snout to vent -SVL), body weight, developmental stage, asynchronous development, and thyroid histopathology. Of these endpoints, SVL and body weight are measures of growth, whereas developmental stage, asynchronous development, HLL, and thyroid histopathology are in the assessment of thyroid axis disruption. Recently, the relevance of suitability of hindlimb length normalized to SVL as a marker of thyroid disruption has been questioned based on its relationship to the growth endpoints (SVL, weight) and the relationship between limb length and differentiation. To evaluate normalized HLL, the correlation between HLL and either SVL or body weight was evaluated in the controls from 10 independently performed AMA studies at study day (SD) 21. Eight of the 10 AMA studies did not have significant late stage development per OECD Test No. 231 and USEPA Test Guidelines OPPTS 890.1100. For the 2 studies, data were censored to separate ≥ NF stage 60 from the >NF stage 60. Negative or no correlation between hindlimb length and SVL was found in 7 of the 8 studies examined without late stage development (r² = 0.315-0.275, 0.553). Negative or no correlation between HLL and SVL length of 6 and 12, respectively, the correlation between hindlimb stage development (r² = 0.347-0.156, 0.429, 0.564). For the censored studies, correction between HLL and SVL or body weight was found in 1 of the 2 studies (r² = 0.452, 0.511). In each of the 10 studies, no asynchronous development was consistent with the absence histopathological findings in the control. The degree of HLL differentiation relative to other morphological markers of developmental stage determination if asynchronous development occurred. Since hindlimb differentiation is controlled by the thyroid axis during metamorphosis, it represents a more suitable endpoint in assessing potential thyroid disruption. To conclude, hindlimb differentiation, developmental stage and thyroid histopathology should be used in a weight-of-evidence based assessment of thyroid axis disruption. Normalized HLL should not be included in the assessment.

**WE381**

Acute toxicity testing using Mediterranean fish species (Dicentrarchus labrax L., 1758): Intercalibration exercises towards standardized procedure
Diatoms are not only part of the risk assessment (RA) for plant protection products, photosynthetically ac. According to OECD Test Guideline 201, the importance of a continuous improvement of the technical setup for a successful performance of ecotoxicological test. The addi. In this presentation several examples for the testing of aquatic toxicity testing of difficult substances and mixtures (23) provides some hints how the “standard testing” with these difficult tests can be conducted, but due to the numberless combinations of characteristics of these difficult substances, some innovation is required to find the best test design for the individual chemicals. We show examples for the toxicity testing of difficult test items starting with the investigation of the characteristics of the test item in the respective test water (water solubility, stability in water, photolysis effect, adsorption, storage conditions). The new Technology evaluating Acartia tonsa as a biological model to be used on ecotoxicological studies or live food for larviculture. On of the particular, the lack of specific acute toxicity methods on Mediterranean fish species have involved the adaptation of procedures available for freshwater fish (OECD, 203, 1992). In order to standardize the acute toxicity method for European sea bass (Dicentrarchus labrax L., 1758) larvae (species widespread in Mediterranean sea), two intercalibration exercises were conducted by 7 Italian laboratories, according to ISO/IEC 43-2:1997 and ILAC-G13:2000. To this end, for every exercise, the laboratories were instructed to use the same test protocol for the first (interlaboratory variability of the tests were verified and Z-scores computed; the intra and inter laboratory variability of the tests were verified and Z-scores computed). Statistical analyses showed no significant differences in the data produced by most of the laboratories. The results indicate the standardization procedure is in advanced stage.

We382 Introduction of a New Dosing System for Chronic Fish Tests Conducted with Difficult Substances S. Höger, Innovative Environmental Services (IES) Ltd / Environmental Toxicology; A. Peither, J. Schreitmüller, Innovative Environmental Services IES Ltd Chronic toxicity tests with fish are required for the risk assessment of plant protection products, pharmaceuticals and chemicals (depending on the damage and characteristics of the chemical). As chronic standard toxicity test the fish Early Life Stage (ELS) test following the OECD Guideline (GL 210) has to be conducted, in certain cases this test system can be extended to an OECD GL 229, 230, 234 or to a full life cycle test. All these tests include the evaluation of sublethal effects on the test fish. This extension is recommended in case an influence of the substance towards the endocrine system is known. The following 2 (interlaboratory variability of the tests were verified and Z-scores computed; the intra and inter laboratory variability of the tests were verified and Z-scores computed). Statistical analyses showed no significant differences in the data produced by most of the laboratories. The results indicate the standardization procedure is in advanced stage.

We383 Difficult Substances as Challenge for the Algal Growth Inhibition Test According to OECD Test Guideline 201 S. Höger, Innovative Environmental Services (IES) Ltd / Environmental Toxicology; A. Dupont, J. Schreitmüller, Innovative Environmental Services IES Ltd Photosynthetically active organisms such as green algae, blue green algae and diatoms are not only part of the risk assessment (RA) for plant protection products, but also standard test organisms for the RA for pharmaceuticals and chemicals (REACH). Especially chemicals as basic element for the synthesis of more complex products show a broad variety of characteristics from well water soluble, stable and non-toxic to hardly water soluble, unstable, volatile and toxic for water organisms. The group of chemicals with one or more of the latter characteristics is a challenge for the toxicity test with aquatic organisms. The OECD guidance document on aquatic toxicity testing of difficult substances and mixtures (23) provides some hints how the “standard testing” with these difficult tests can be conducted, but due to the numberless combinations of characteristics of these difficult substances, some innovation is required to find the best test design for the individual chemicals. We show examples for the toxicity testing of difficult test items starting with the investigation of the characteristics of the test item in the respective test water (water solubility, stability in water, photolysis effect, adsorption, storage conditions). The new Technology evaluating Acartia tonsa as a biological model to be used on ecotoxicological studies or live food for larviculture. On of the particular, the lack of specific acute toxicity methods on Mediterranean fish species have involved the adaptation of procedures available for freshwater fish (OECD, 203, 1992). In order to standardize the acute toxicity method for European sea bass (Dicentrarchus labrax L., 1758) larvae (species widespread in Mediterranean sea), two intercalibration exercises were conducted by 7 Italian laboratories, according to ISO/IEC 43-2:1997 and ILAC-G13:2000. To this end, for every exercise, the laboratories were instructed to use the same test protocol for the first (interlaboratory variability of the tests were verified and Z-scores computed; the intra and inter laboratory variability of the tests were verified and Z-scores computed). Statistical analyses showed no significant differences in the data produced by most of the laboratories. The results indicate the standardization procedure is in advanced stage.

We384 Activity based Collombola sampling may improve the data of field studies for regulatory purposes P. Mack, A. Appeltauer, J. Illig, Eurofins Agrofins Services Ecotoc GmbH; S. Knaebe, EAS Ecotoc GmbH / Ecotoc Field Soil micro-arthropod field studies are carried out as part of the risk assessment of plant protection products. Those studies followed the proposed study design for soil organisms by Römbke et al. (2009). Soil cores are taken in the field and afterwards soil organisms are extracted from the soil using high gradient extraction. Until now little is known about vertical movements of collombolans. Especially in long periods with high temperatures and low precipitation, a high number of collombolans might mitigate in deeper soil layers as included in the standard sampling scenario of 5 - 10 cm soil cores. Therefore it might be useful to cover also deeper soil layers, which contain potentially more specimens at the sampling time. One activity based trapping method for soil microarthropods would be the side traps which were presented at SETAC 2016 by Dehelen et al. 2015. Our poster will discuss possible advantages for the combination of soil core and slide trap sampling and will present first results from the comparison of soil core and slide trap catches. Römbke, J., Schmelz, R., Knaebe, S., 2009: Field studies for the assessment of pesticides with soil mesofauna, in particular enchytraeids, mites and nematodes: Design and first results. Soil Organisms, 81: 237-264 Stefan-Bogdan Dehelen et al., 2016 Stratification of soil arthropods in topsoil layers, SETAC Europe 26th Annual Meeting, Nantes, France

We385 New Technology evaluating Acartia tonsa as a biological model S. Abeur, University of Aveiro / Dep. Biology & CESAM; S.M. Leandro, Polytechnic Institute of Leiria / MARE Marine and Environmental Sciences Center, A.M. Soares, University of Aveiro / department of Biology & CESAM; R.E. Martins, M. Oliveira e Silva, University of Aveiro / DETI IEETA Copepods play an important ecological role on marine ecosystems and may act as a sentinel of environmental degradation resulting from direct or indirect human impacts. The copepod Acartia tonsa is a calanoid species with a worldwide distribution and relatively easy to maintain for several generations under captive conditions. These characteristics allow this species to be a potential biological model to be used on ecotoxicological studies or live food for larviculture. On of the bottlenecks for its massive utilization relies on the time consumption procedures related with counting and cultures monitoring. To overcome such constrain, the aim of the present study was to evaluate the application feasibility of a new technology based on automatic counting and counting which can be used on live food and particles and/or organisms. The technology D Counter constitutes an innovative approach, by the fact of turning the data harvesting process much more efficient and accurate, breaking the traditional, error-prone, human-based counting methodology. The obtained results for A. tonsa cultures indicates a high significant correlation between manual and automatic counting, constituting the first step for the use of this biological model on experimental studies.

We386 Solubility limits of lanthanides in standardized ecotoxicological media
WE387
Improving ecotoxicity tests for trace elements forming poorly soluble chemical species in test media
D.A. Vignati, CNRS / LIEC UMR7360; F.C. Acanfora, University of Salerno / Department of Chemical and Biology; G. Cornelis, Swedish University of Agricultural Sciences / Soil and environment; M. Carotenuto, University of Salerno / Department of Chemical and Biology; B.J. Ferrari, Centre Ecotoxic EAWGPEP; G. Lofrano, University of Salerno / Department of Chemical and Biology
In order to get an appropriate interpretation of ecotoxicological results the exposure concentrations of test organisms to the contaminant of concern must be kept constant and stable over the test duration. Increasing evidence suggests that this is often not the case when dealing with elements that tend to form chemical species with low solubility (e.g., oxides and oxyhydroxides for Cr(III) and Sn phosphates in the case of some lanthanides). In such situations, the calculation of meaningful effect concentrations for hazard and risk assessment must consider the temporal decrease of exposure concentrations to avoid erroneous conclusions. We performed ecotoxicity tests using semistatic exposure conditions to assess if periodical medium renewal could compensate for the temporal decrease in element concentrations. While thermodynamic modeling does not account for the kinetics aspects of speciation, it still provides useful indications as to the actual exposure conditions likely to be experienced by organisms in standard ecotoxicity tests. As limonene is a narcotic substance its Chronic 1 assignment was expected to be classified under category 1 in accordance with the ECSTAC 4th Edition guidelines. However, results for the acute and chronic tests did not support such an assignment, which equates to chronic category 1 classification is extrapolated from the acute category 1 toxicity, log Kow >4 and erroneously assumed not-rapid biodegradability of the substance. As limonene is a narcotic substance its Chronic 1 assignment was expected to be conservative. Due to doubts on the chronic classification and the consequences for labelling, storage, handling and transport, long term aquatic ecotoxicity studies were conducted to obtain a solid basis for the environmental classification. In this context, testing LNs ecotoxicity following established standard protocols must consider the peculiar chemistry of these elements if meaningful results are to be obtained and used to establish regulatory limits. After addition to ecotoxicological media, typically in the form of soluble chloride salts, LNs can rapidly form complexes with phosphates or carboxylates or undergo hydrolysis. Due to the low solubility of LN-phosphates and LN-carboxylates their exposure may be the formation and solubility of LNs in test media. The actual exposure conditions likely to be experienced by organisms in standard ecotoxicity tests will further complicate the interpretation of the corresponding biological responses. In this contribution, we use thermodynamic speciation modeling to examine the equilibrium theoretical speciation of LN in standardized ecotoxicological media for algae and invertebrates, and in the environment for the specific case of chromium. The results showed that when tested in published laboratory studies, attempts are also made to model the fraction of added LN predicted to occur in precipitated forms. Finally, depending on the availability of the appropriate complexation constants, the predictions for laboratory media are compared with those for natural waters where the presence of dissolved organic matter can cause important differences in speciation with laboratory media. In such situations, the calculation of meaningful effect concentrations for hazard and risk assessment must consider the temporal decrease of exposure concentrations to avoid erroneous conclusions. We performed ecotoxicity tests using semistatic exposure conditions to assess if periodical medium renewal could compensate for the temporal decrease in element concentrations. While thermodynamic modeling does not account for the kinetics aspects of speciation, it still provides useful indications as to the actual exposure conditions likely to be experienced by organisms in standard ecotoxicity tests.

WE388
Long term ecotoxicity testing of limonene for hazard classification: not such a lemon after all
P.C. Thomas, CEHTRA SAS / Ecotoxicology and Risk Assessment; F. Balk, Royal Haskoning DHV; H. van Bergen, Para-Celsus concept; K. Jenner, Givaudan /
Global Regulatory Affairs & Product Safety; A. Kamper, DHI; S. Gimeno, Firmenich / Product Safety and Regulatory Affairs
Limonene is a stereosymmetric substance taking its name from lemon, which dominates the composition of the essential oils of citrus fruits. When synthesised as dipentene it is a racemate. However, in botanical sources it is present as the D- or L-enantiomer in Natural Complex Substances (NCSs) obtained at anything from traces in some plant extracts to over ninety percent in orange oil. These oils are commercially used as fragrance and / or flavour ingredients. They are naturally present in many products (cosmetic products, food manufacture, fragrance perfumery, botanical insecticide, household cleaning products, etc.). Therefore, it is predominantly released back to the environment after use. The racemate and both D- and L-limonene received a harmonised classification under Annex VI of the EU C&L legislation as: "Aquatic acute 3 (Very toxic to aquatic life) and Aquatic chronic category 1: (Very toxic to aquatic life with long-lasting effects). Both classifications have a severe impact on storage, handling and transport requirements of limonene and the many (natural) complex substances and fragrance and flavour mixtures that contain even small amounts of it. The existing classification is a result of a limited data set notably for chronic endpoints. The chronic category 1 classification is extrapolated from the acute category 1 toxicity, log Kow >4 and erroneously assumed not-rapid biodegradability of the substance. In limonene is a narcotic substance its Chronic 1 assignment was expected to be conservative. Due to doubts on the chronic classification and the consequences for labelling, storage, handling and transport, long term aquatic ecotoxicity studies were conducted to obtain a solid basis for the environmental classification. Log Kow >4 and erroneously assumed not-rapid biodegradability of the substance. 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Due to doubts on the chronic classification and the consequences for labelling, storage, handling and transport, long term aquatic ecotoxicity studies were conducted to obtain a solid basis for the environmental classification. Log Kow >4 and erroneously assumed not-rapid biodegradability of the substance. In this context, testing LNs ecotoxicity following established standard protocols must consider the peculiar chemistry of these elements if meaningful results are to be obtained and used to establish regulatory limits. After addition to ecotoxicological media, typically in the form of soluble chloride salts, LNs can rapidly form complexes with phosphates or carboxylates or undergo hydrolysis. Due to the low solubility of LN-phosphates and LN-carboxylates their exposure may be the formation and solubility of LNs in test media. The actual exposure conditions likely to be experienced by organisms in standard ecotoxicity tests will further complicate the interpretation of the corresponding biological responses. In this contribution, we use thermodynamic speciation modeling to examine the equilibrium theoretical speciation of LN in standardized ecotoxicological media for algae and invertebrates, and in the environment for the specific case of chromium. The results showed that when tested in published laboratory studies, attempts are also made to model the fraction of added LN predicted to occur in precipitated forms. Finally, depending on the availability of the appropriate complexation constants, the predictions for laboratory media are compared with those for natural waters where the presence of dissolved organic matter can cause important differences in speciation with laboratory media. In such situations, the calculation of meaningful effect concentrations for hazard and risk assessment must consider the temporal decrease of exposure concentrations to avoid erroneous conclusions. We performed ecotoxicity tests using semistatic exposure conditions to assess if periodical medium renewal could compensate for the temporal decrease in element concentrations. While thermodynamic modeling does not account for the kinetics aspects of speciation, it still provides useful indications as to the actual exposure conditions likely to be experienced by organisms in standard ecotoxicity tests.
used as medicinal products, e.g. gene therapies, into the environment is regulated by directive 2001/18/EC of the European Parliament and of the Council of 12 March 2001. An integral part of the directive regulates the provision of information on the GMO and, based on this, the risk management with regard to the environmental effects of such releases. As regulated by this directive, a publicly accessible database (the “GMO Register” of the JOINT RESEARCH CENTER of the EC (http://gmoinfo.jrc.ec.europa.eu)) contains information about all GMOs under study. As of 07.11.2016, there were 238 entries of medicinal GMOs in the “Summary Notification Information Format (SNIF).” SNIFs are prepared as a summary document of the confidential environmental risk assessments (ERA) by the respective Sponsors of clinical trials in the EU and evaluated during the clinical trial application by the national competent authorities. These trials, inter alia, information regarding the GMOs and the parental organism’s nature, release, environmental interactions, monitoring, waste treatment and emergency response plans. We strive to assess information concerning the environmental risk, derived measures and the overall standard of SNIFs concerning compliance with the regulatory requirements. To do so, we picked a homogeneous group of GMOs, namely gene modified Aedes virus, the most frequently used vector in gene therapy trials worldwide. Relevant information were entered into a database and categorized, applying unified vocabulary. Different challenges regarding the information available within the SNIFs were identified by analyzing the database: in several cases mandatory information was not available, e.g. monitoring plans, and in other cases the SNIF documents were misinterpreted, e.g. the connection between replication, dissemination and survivability was interpreted here as different concepts of PBT/vPvB property. Unpredictable effects are fundamentally linked to uncertainty in: Assessing accurately a chemical’s potential to amplify along the food chain, and Whether a chronic value can be established based on its MoA. Significant scientific progress has been made in the field of aquatic toxicity testing of difficult chemicals, evaluation and interpretation of ecotoxicological data since the PBT/vPvB criteria are determined, applied and categorized, applying unified vocabulary. Differently, it is proposed to specify some parts of the SNIFs in order to make more reliable information transparently available.

**WE391**

**PBT evaluation 20 years on: is it time to reconsider the technical progress made in risk assessment methodology?**

P. Thomas, C. Durou, CETHRA SAS

In the EU, the ecotoxicological dataset for a chemical is used for the purposes of prospective risk assessment (PRA) and of PBT assessment. While the PRA aims at determining the use conditions and risk management for which the environment is safe, the PBT assessment aims at identifying chemical for which effects on the long-term are considered unpredictable and that environmental exposure is difficult to reverse. The scope of this poster is to discuss, in the light of technical and scientific progress: For which PBT-Like and certain PBT chemical, PRA can now be carried out The justification of the numerical criteria behind the identification of PBT/vPvB property Unpredictable effects are fundamentally linked to uncertainty in: Assessing accurately a chemical’s potential to amplify along the food chain, and Whether a chronic value can be established based on its MoA. Significant scientific progress has been made in the field of aquatic toxicity testing of difficult chemicals, evaluation and interpretation of ecotoxicological data since the PBT/vPvB criteria were originally designated. The numerical criteria were established in the late 1990s by OSPAR. Although the primary basis has been transformed environments and used by the EU (TC NES) from the early 2000’s enlarging the protection goal to any environment. The criteria became applicable to any chemical produced over 10 TPA when REACh regulation came into force, thus, treating chemicals as a homogeneous group. Interestingly, the criteria were originally defined by using data from a set of chemicals known as highly hazardous for the environment. Such compounds (e.g., aromatic compounds) were non-toxic and hydrophobic while the chemical space is much more diverse. In other words, a BCF value of 2000 may be a good cut-off for chemicals which are both highly hydrophobic and slowly metabolised to anticipate amplification along a food chain but may be of limited meaning for other chemical classes. In the US, B is defined as a BCF of > 5000 which is the B criterion in the EU while perhaps the only meaningful way to determine B is to consider bioaccumulation in the food chain which has no legal relationship with the B criterion. Further questions can be posed of the true meaning of the half-life cut-off values for P and vP in terms of environmental persistence and the meaningfulness of using a standard mg/L cut off basis for T blanketling all MoAs.

**WE392**

**UCVB block method for estimating expected mixture toxic pressure of substances of Unknown or Variable composition, Complex reaction products or Biological materials**

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We have developed a spreadsheet calculation tool for chemical safety assessment of UCVB substances. The tools adopts the approach of Concave’s Hydrocarbon Block Method for chemical safety assessment of complex petroleum substances. The tool is meant to be used for demonstrating ‘safe use’ of chemicals, as required for registration of substances under REACH. The tool makes use of scientifically up to date versions of the exposure models SimpleTreat and SimpleBox, combined with state-of-art expected mixture pressure calculation using the Van Straalen-Aldenberg convolution integral. The tool estimates the addition of toxic pressure to the ambient toxic pressures in local, regional and continental EU environments, expected from the UVCB mixture. It is proposed that ‘safe use’ is demonstrated sufficiently well by showing that the UVCB under study is expected to contribute negligibly to ambient toxic pressure in the environment. The proposed ‘safe use’ calculation method has been tested in an extensively well studied UVCBs. In the poster briefly explains the new UVCB block method and illustrates its potential with the outcomes of test calculations.

**WE393**

**Evaluation of hypopharyngeal glands development in Honeybees (Apis mellifera L.) from toxicity studies in the light of current guidelines (EFSA and OECD)**


Honey Bee (Apis mellifera L.) is a species that belongs to a group called ‘beneficial insects’. All arthropods from this group play the important roles in nature, albeit bees go a few steps forward and they also find application in the food, pharmaceutical and others industries. With the current decline in bee’s colony numbers, these arthropods should be handled with extreme caution. Therefore, it is extremely important to assess the risk for non-target organisms for which plant protection products are often more toxic than pests of agricultural crops. This assessment is related not only to the direct influence of chemicals on the number of bee populations, causing morphological mortality or morbidity, but also indirectly - through the impairment of the ability to raise the larvae, for example by disturbing the work of hypopharyngeal glands (HPG) responsible for the production of ‘milk’ containing proteinic substances to feed larvae and queen. By 2017, the only document regulating the toxicity study of chemicals on bees was the EFSA document (EFSA Journal 2013;11(7):3295), which included continuous access to distilled water and pollen and evaluation of HPG. However, in 2017, the new OECD guideline (no. 245) was introduced, in which the methodology of chronic toxicity testing was changed compared to the EFSA document. Changes occurred in the way of dealing with bees – there is no access to distilled water and pollen, and no evaluation of HPG. It is a significant change, because according to the literature for several species of hypopharyngeal glands do not develop correctly in these conditions, which exclude the assessment of HPG. Hence OECD guideline probably does not consider it as endpoint in the study. As previous study has shown, chemicals can have influence on development of hypopharyngeal glands, without causing mortality and morbidity. This matter is worthy considering and should be investigated further, in order to introduce the evaluation of hypopharyngeal glands as an endpoint in toxicity testing of chemicals on bees.

**WE394**

**Assessing toxicity to Daphnia magna using movement parameters**

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*Overview* The numerous studies, hypopharyngeal glands do not develop correctly in these conditions, which exclude the assessment of HPG. Hence OECD guideline probably does not consider it as endpoint in the study. As previous study has shown, chemicals can have influence on development of hypopharyngeal glands, without causing mortality and morbidity. This matter is worthy considering and should be investigated further, in order to introduce the evaluation of hypopharyngeal glands as an endpoint in toxicity testing of chemicals on bees.

**WE395**

**The validation of analytical methods in ecotoxicology**

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The validation of analytical methods (regulated by SANCO/3029/99 rev 4.) used in support of ecotoxicological studies has become an important aspect of the
A new pulsed-exposure early life stage test design for rainbow trout on an insecticide. Refining OECD Guideline 210 to meet the needs of EFSA Aquatic Guidance 2013

C.S. Ramsden, AgroChemex Environmental Ltd / School of Biomedical and Biological Sciences; C. Gamblin, AgroChemex Environmental Ltd; W.R. Jenkins, W R Jenkins / Regulatory Affairs Ecotoxicology; S. Norman, RidgewayEco

Constant-exposure in OECD TG 210 Fish Early Life Stage studies is unrealistic for fast-dissipating pesticides compared to edge-of-field water-bodies. EFSA Aquatic Guidance (2013) allows aquatic toxicity studies to be modified so the exposure-profile (peak-height, pulse-duration, number of pulses) is comparable to the worst-case predicted field-exposure. In the present study on a synthetic pyrethroid (SP) insecticide, a novel method was developed to simultaneously assess effects on 3 early-life stages of rainbow trout. The TG 210 design was modified to incorporate a worst-case time-variable exposure profile in tanks containing a 10 mm sediment layer and stainless steel mesh barrier to allow water movement but prevent fish disturbing the sediment. Life stages used were newly fertilised 'eggs', 'alevins' (non-feeding larvae) and free-feeding 'swim-up' fry. To ensure physical separation of the 3 life stages within the tank, eggs and alevins were each held in a glass incubation tube with a mesh base. A control group plus 5 concentrations were used. To start, each group had 4 replicate tanks each with 50 eggs, 20 alevins and 20 swim-up fry. The study duration for organisms starting as 'eggs', 'alevins' and 'swim-up' fry was 72, 45 and 31 days respectively. This allowed for the assessment of effects over a period including at least 2 weeks of growth after initiation of free-feeding for each of the 3 life stages. Standard end points were assessed including hatch success, survival, growth and clinical signs (e.g. loss of equilibrium and coordination). To assess the potential neurotoxic action feeding behaviour was categorised as active, passive and not feeding. This refined-exposure study showed that 3 critical life stages of fish can be tested simultaneously, whilst complying with the fundamental elements of TG 210. Control hatch and survival rates were both > 95%. The design allowed the direct comparison of the sensitivity of each life stage to the same concentrations and profiles. Free-feeding 'swim-up' fry was the most sensitive exposed life stage, based on clinical signs, feeding and slightly reduced growth. Swim-up of exposed alevins was delayed at high treatment levels. Exposed eggs were unaffected.

Distribution, transformations and biological effects of incidental nanoparticles and nanostructures in the environment from a current point of view (P)

WE397

Dissolution of Different Silica Nanoparticles in Aqueous Matrices


Since centuries, silica (SiO₂) is used in large scale industrial applications, such as cement manufacture or glass production. In these applications, SiO₂ is used in its bulk form. Recently, SiO₂ in nanoparticulate form has broadened the range of applications, both in large scale, e.g. as anti-cake agent in food industry, or owing to its abrasive effect in cosmetics; and in small scale, for the production of biosensors, swim-up fry. There were two 22 hour stages [1]. The use of silica nanoparticles (SiO₂-NPs) as a delivery vehicle for molecules in plants is being investigated [2], demonstrating their potential in environmental applications. Many of the current uses increase the likelihood that SiO₂-NPs could accumulate in the environment and in food webs. Therefore, it is crucial to investigate the dissolution of SiO₂-NPs in different environments. Besides information about the degradability of the nanomaterials, this knowledge will also allow to produce more environmentally friendly products. Here, we present the results of a method development to quantify the dissolution of different SiO₂-NPs in aqueous media in order to determine the dissolution kinetics. This method relies on the use of inductively coupled plasma optical emission spectroscopy (ICP-OES) for the quantification and the parallel characterization of the particles by transmission electron microscopy (TEM) and dynamic light scattering (DLS). A simple setup based on dialysis membranes, and a sampling protocol are in the process of being established. The first results indicate that is possible to detect the dissoluted fraction of SiO₂-NPs roughly 60 nm in diameter within about one day. Long-term dissolution experiments will be performed in the coming months to determine the dissolution kinetics more precisely. This setup will serve as a tool to assess the behavior of SiO₂-NPs in environmental media. [1] Barki TK, Sahu B, Swain V. 2008. Nanosilica—from medicine to pest control. Parasitology Research. 103:253, [2] Torney F, Trewyn BG, Lin VSY, Wang K. 2007. Mesoporous silica nanoparticles deliver DNA and chemicals into plants. Nature Nanotechnology. 2:295. Acknowledgement - The authors thank the Swiss National Science Foundation (project 168187) and the Adolphe Merkle Foundation for the support and funding of the study, and Laura Rodriguez-Lorenzo for her precious advice and suggestions.

WE398

Occurrence of fullerene aggregates in Mediterranean rivers: Two cases of study

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Fullerenes are carbon nanostructures that have many potential applications in nanotechnology and that can be generated in some combustion processes. Since their environmental effects and behaviour are uncertain, fullerenes are considered emerging contaminants and during the last years they have been included in some aerial and terrestrial studies [1-3]. However, few of these studies have reported the presence of fullerene in water systems. In order to assess the environmental risk of fullerenes it is important to enlarge the dataset and to characterize the concentrations of these nanomaterials in different rivers. In the present work, C₆₀ fullerenes, C₇₀ fullerenes and five functionalized derivatives were determined in water and sediment samples from two Mediterranean rivers. The first case of study was located in the Sava River (Soutern Europe), where more than 30 samples were studied in two sampling campaigns. In the second case of study, samples of estuary water, wastewater, river water and coastal water from the Ebro River delta were analysed. In both studies, C₆₀ was the most ubiquitous compound and it was detected for the first time in the marine environment, although its concentrations were below the ng/l order in all the cases. The toxic fullerene 

WE399

Occurrence, fate and behaviour of fullerenes in the environment

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The occurrence, fate and behaviour of carbon nanomaterials in the aquatic environment are dominated by their functionalization, association with organic material and aggregation behaviour. In particular, the degradation of fullerene aggregates in the aquatic environment is a primary influence on their mobility, sorption potential and toxicity. In this presentation, a summary of the occurrence of fullerenes in environmental matrices performed in different studies of our group will be presented. The analytical approach to investigate seven fullerenes (C₆₀, C₇₀, N-methylfulleropyrroline, [6,6]-phenyl C₆₀ butyric acid methyl ester, [6,6]-thienyl C₆₀ butyric acid methyl ester, C₆₀ pyrrolidine tris-acid ethyl ester and [6,6]-phenyl C₆₀ butyric acid methyl ester) in waters, soils and sediments combines an on-line derivatization-assisted separation (dSPE, UHPLC) followed by liquid chromatography (LC), using a pyrenyl[propyl] group bonded silica based column, coupled to a high-resolution mass spectrometer (HRMS) using atmospheric pressure photo ionisation (APPI) in negative ion mode. Main results of these studies showed levels of pg/m²-ng/m² in atmospheric aerosols, pg/g-ng/g in soils and pg/l-ng/l in river waters. The composition of different fullerenes including pristine fullerenes (C₆₀ and C₇₀) and functionalized ones from the engineered origin will be discussed. In addition, different degradation studies of fullerenes in water suspensions emulating different environmental conditions and during a wastewater treatment will be presented. Degradation studies have been carried out under controlled conditions of salinity, the humic substances content, the pH and the sunlight irradiation. The results of degradation studies will show that up to ten transformation products are produced, including epoxides and dimers. Finally, the kinetics of generation of each transformation product will be well presented.

WE400
The influence of engineered surface coatings on nanomaterial stability in a complex, natural medium

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The colloidal stability of engineered nanomaterials (ENMs) within aquatic environments has been shown to be highly sensitive to the particular ENM surface coating. Research using simplified, synthetic mediums has demonstrated that ENM stability is strongly influenced by the engineered surface coating enveloping the ENM. However, it is uncertain whether this coating will continue to dictate particle stability when the ENM is dispersed within a complex, natural medium. Instead, it has been suggested that the “eco-corona” acquired by the ENM via interactions with the surrounding environment will govern ENM stability. This implies that within these mediums, engineered surface coatings will have a negligible impact upon ENM stability. The aim of this research was to investigate this subject further and determine whether an ENMs’ engineered surface coating remains a relevant factor effecting ENM stability in a complex, natural medium. In Using samples of a local freshwater river to represent a complex, natural medium, a suite of batch experiments were conducted. Each batch was dosed with a single model ENM, which included 12-15 nm gold-core nanoparticles (AuNPs) with different surface coatings (e.g., diverse surface charges, stabilization mechanisms). Aliquots were collected from each batch over time, immediately centrifuged to remove large particles/aggregates, and the supernatant collected for analysis via ICP-OES. From this, the concentration of unaggregated AuNPs remaining in the supernatant over the overall contact time was measured. As the mixing制度, we will see increased input of ENPs into WWTPs. Therefore the increased use and potential toxicity of ENPs poses a challenge for WWTPs due to their potential harmful effects towards activated sludge. The extent to which WWTPs can remove ENPs from the sewage must also be explored, to determine not only likely outflow into receiving waters but also accumulation within the activated sludge itself. To this end we focused on the activated sludge treatment, as the majority of ENPs can remain in wastewater stream throughout preliminary and primary stages, we investigated a range of ENP digestion and analysis protocols to determine the most reliable procedure for ENP analysis from activated sludge. From this, we developed an analytical method involving H2SO4-HNO3 microwave assisted digestion coupled with ICP-OES to measure ENP concentrations. Following this, using laboratory microcosms we assessed the kinetics of ENP removal by activated sludge. The kinetic design we adopted provided different ENPs-activated sludge contact times. ENP concentrations were then analysed in both effluent and settled fraction. Similarly to previous reports, high and quick TiO2 removal rate (>80% and >99% after respectively 5 and 60 min) during activated stage treatment have been found. However, the dependency of ENPs involved in the removal of ENPs from wastewater by activated sludge remain not fully understood, but results of laboratory test and site samples indicate ENPs are rapidly captured-associated with activated sludge. Following this, we exposed activated sludge to repeated cycles of ENP exposure indicative of the cycling of activated sludge in a WWTP. During each cycle, ENPs were efficiently removed. As consequence of this exposure, the significant enrichment of activated sludge biomass with metal based ENPs can result in a secondary hazard, as ENPs rich biomass acting as a “sponge” can accumulate high concentrations of ENPs, which may be released when recirculated within the wastewater treatment or when applied to land. A similar approach is now being adopted to investigate the fate of mixture of ENPs and ENPs from real products within the activated sludge treatment.

WE402 Fate factor of engineered TiO2 nanoparticles in aquatic and terrestrial natural environments

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Nanoparticles are defined as nano-objects between 1 and 100 nanometers in size. Engineered TiO2 nanoparticles are used in several fields such as construction, cosmetic and food which leads to an important production and inevitably to emissions generating environmental impacts. To quantify them, the Life Cycle Assessment is a powerful method that is able to characterize TiO2 NPs according to their fate in environmental media and their effects on ecosystems and human health. The main objective of this study is to determine the engineered TiO2 nanoparticles (TiO2 ENPs) fate according to two approaches: experimental and by calculation. For this purpose, it requires firstly to detect and quantify TiO2 NPs in water, soil and sediments near a production site in Vieux-Thann (68) to determine parameters which contribution on TiO2 NPs and several environmental factors. ENM stability is influenced by the engineered surface coating, even after interacting with a complex, natural medium.

WE403 Assessing the fate and transport of engineered TiO2 nanoparticles in sewer pipes through a dynamic multimedia model (SWNano)

K. Kim, Seoul National University / Environmental Planning Institute Graduate School of Environmental Studies

During the past decades, the nanotechnology and use of engineered nanoparticles (ENPs) have rapidly developed. It is therefore inevitable that they enter the natural environment such as rivers and marine environments. So various models have developed to expect exposure amounts in environment for proper risk assessment of ENPs. For that purpose there is a need for a powerful method, called Environmental Fate Assessmentis a powerful method that is able to characterize TiO2 NPs according to their fate in environmental media and their effects on ecosystems and human health. The main objective of this study is to determine the engineered TiO2 nanoparticles (TiO2 ENPs) fate according to two approaches: experimental and by calculation. For this purpose, it requires firstly to detect and quantify TiO2 NPs in water, soil and sediments near a production site in Vieux-Thann (68) to determine parameters which contribution on TiO2 NPs and several environmental factors. ENM stability is influenced by the engineered surface coating, even after interacting with a complex, natural medium.

WE404 The importance of cell wall of marine microalgae in preventing the toxicity of nanoparticles

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Metallic and oxo metal oxide nanoparticles (NPs), such as Au and CeO2 NPs, have increased their global production because they have been widely used in new applications and consumer products such as textile, personal care product, biomedicine and catalysis. NPs-containing wastes discharged in aquatic systems
have produced undesirable effects in many marine organisms. Marine phytoplankton is vital in marine ecosystems, as microalgae are at the bottom of the food web and, therefore, any change in microalgae population will have an important impact into the rest of food web. The direct mechanism of NPs toxicity is the physical damage in cell membrane through adsorption of NPs onto the cell wall leading to NPs uptake, bioaccumulation and toxicity in different organelles. Therefore, the hypothesis of this work is that microalgae lacking of cell wall will be more vulnerable to the acute effects of NPs than those associated with a typical cell wall. To test this hypothesis two microalgae species, Dunaliella salina, lacking cell wall, and Chlorrella autotrophica, with a typical cellulosic cell wall were chosen. Species were exposed to ion (AgNO3 , Cu(NO3)2 ) and NPs (Ag NPs and CeO2 NPs) forms of Ag and Ce over 72 h and the following responses were assessed: cell density, cell viability, cell size, cell complexity, autofluorescence of chlorophyll a, active chlorophyll, effective quantum yield of photosystem II and reactive oxygen species (ROS). Metals in both forms (NPs and ionic) caused negative effects in cell division, inherent cell properties and physiological mechanisms of both microalgae. The general trend was a decrease in active chlorophyll, effective quantum yield of PSII, and cell density and an increase in cell complexity and percentage of intracellular ROS. For both marine species, Ag was more toxic than Ce and ionic forms of both metals were more toxic than NPs. Contrarily to our hypothesis, D. salina, despite not having a cell wall, showed to be less sensitive to metals than C. autotrophica. Therefore, the cell wall of C. autotrophica seems not to suppose higher protection preventing toxicity of NPs. The higher resistance of D. salina against the metals and metallic NPs tested might be related to: (i) its ability to stock-displications, the measured z-average ranged from 600 nm (CPO-27-Ni) to 8 μm (HKUST, Zn-CPO, FeBTC-JM-AR and CPO-27-Ni) are investigated in relevant environmental test media. Furthermore, we study the dissolution of metals and other elements from NPs in test media, and their contribution to the observed effects on R. subcapitata. Particle size measurements showed that the NPs have a primary particle size between 200 nm and several micrometres. In freshly sonicated stock dispersions, the measured z-average ranged from 600 nm (CPO-27-Ni) to 8 μm (HKUST, Zn-CPO, FeBTC-JM-AR and CPO-27-Ni) at 8 μm (HKUST, Zn-CPO, FeBTC-JM-AR and CPO-27-Ni) and Zn-CPO had the highest zeta-potential of -25 and -20 mV respectively, with Al (OH) fumarate and FeBTC-JM-AR bearing a positive surface charge. Uio-66-COOH and HKUST revealed increased susceptibility to ionic Ag, while Uio-66-COOH and HKUST had the most inert material regarding release of dissolved metal due to their adsorption properties, the materials also drastically reduced amount of P in the exposure media, with Uio-66-COOH also decreasing Ca and Mn. Potential mode-of-actions, i.e. impact of NPs particle through depletion of nutrient elements, toxicity of dissolved metals, or effects from organic components will be identified in the R. subcapitata toxicity tests.

WE405 Environmental screening of structured hybrid nanoporous materials developed for industrial adsorption applications A. Booth, J. Farkas, SINTEF Ocean / Environmental Technology; R. Blom, SINTEF Materials and Chemistry Inorganic-organic hybrid nanoporous materials (NPMs), such as amorphous mesoporous aluminosilicates and Metal-Organic Frameworks (MOFs) are designed and developed for numerous applications including health care, industrial cooling systems, air purification and gas storage. Their usage and production is expected to increase significantly within the next years, with new applications such as carbon capture and storage becoming increasingly important. In this study, we investigate the environmental behaviour of 6 NPMs and determine their toxicity and the potential mode-of-actions, i.e. impact of NPs particle through depletion of nutrient elements, toxicity of dissolved metals, or effects from organic components will be identified in the R. subcapitata toxicity tests.

WE406 Tracking Physicochemical Changes of PAHs in the Presence of TiO2 Nanoparticles by Assessment of Biological Responses L. St Mary, Heriot-Watt University / EGIS; D. Patsiou, Heriot Watt University / Environmental Sciences; T. B. Henry, Heriot-Watt University / School of Engineering – Physical Sciences; T. B. Henry, Heriot-Watt University / School of Energy, Geoscience, Infrastructure and Society Polycyclic aromatic hydrocarbons (PAHs) are a class of persistent environmental contaminants with diverse physicochemistry and toxicity. In contaminated aquatic environments, PAHs frequently accumulate in sediments or are sorbed to particulates in the aqueous phase. Some PAHs are phototoxic and have a photo-induced toxicity, but little is known about interactions between PAH photoactivity, sorption, environmental fate, and toxicity. Engineered nanoparticles (NPs) can behave as particle agglomerates that participate in sorption/desorption reactions in the aqueous phase, and some NPs (e.g., TiO2-NPs) also have phototoxicity. Aqueous-phase interactions between PAHs and TiO2-NPs are of interest because they are becoming more environmentally relevant (i.e., as NPs are increasingly released into the environment), and because investigations of sorption/desorption processes, in the context of photoactivation, can provide important new information on physicochemistry of both PAHs and NPs. Previous work conducted by our research group have shown that environment of PAHs onto photo-active NPs promotes photo-catalysis of PAHs thus altering PAHs bioavailability and toxicity under UV-A radiation. In these experiments, bioavailability (cytotoxicity P4501A cytochrome gene expression in larval zebrafish) is used as an analytical tool to demonstrate sorption of anthracene and benzo(a)pyrene to NPs in water. Our objective is to investigate PAH-TiO2-NP sorption under UV-A and visible light and determine the formation of a PAH decomposed by TiO2 photooxidation of polycyclic aromatic hydrocarbons (OPAHs) and their bioactivity. Various combinations of PAH-TiO2-NP preparations will be exposed to UV-A, and changes in gene expression of genes involved in Phase I metabolism (cytochrome P450 cyplal and cyplbl) and Phase II metabolism (gst, epxks; gsh; and epoxide hydrolases ephx1 and ephx2) in early life stages of zebrafish will be assessed. The exploitation of biological responses to investigate changes in PAH and PAH-decomposition effects and product bioavailability during sorption processes will provide novel insight into these processes tested directly within the environmentally relevant aqueous phase.

WE407 Toxicity of TiO2 nanoparticles to freshwater chironomids - pointing out the relevant endpoints D. Šavić Zdravković, Faculty of Sciences and Mathematics, University of Niš / Department of Biology and Ecology; B. Jovanovic, Iowa State University / Natural Resource Ecology and Management; A. Đurđević, J. Stanković, Faculty of Science and Mathematics, University of Niš / Department of Biology and Ecology; D. Mišić, Faculty of Sciences and Mathematics, University of Niš / Department of Biology and Ecology In the environment, nanoplastics are present in a number of chemical forms, exhibiting specific interactions, mobility, biological availability and potential toxicity. Both ecotoxicologists and pathologists have expressed their concerns regarding the potential negative effects of nanoplastics in living systems and the environment. The present study was carried out in order to assess the influence of TiO2 nanoparticles (in the form of human white food colorant E171) on the freshwater midge Chironomus tentans. The safety and consequences of the intake of this form of E171 TiO2 for human health have been recently reconsidered. The experimental design was constructed for the sediment dwelling chironomid larval according to OECD guidelines. Concentrations of 125, 250, 500, 1000, 2000 and 4000 mg E171 TiO2 per kg were tested. The endpoints were considered: mortality, emergence ratio was affected by a higher nanoparticulate TiO2 concentration in the sediment (>1000 mg/kg). Sublethal effects on Chironomus tentans larvae at environmentally relevant concentrations were shown through morphological changes, which were qualified and quantified using the geometric morphometry approach, principal component analysis and canonical variate analysis. This was the first time a geometric morphometric approach was used to assess the deformities in chironomid larvae exposed to nanoplastics. Geometric Morphometrics revealed the tendency of the mentum teeth to narrow and elongate and the mandibles to widen and loose the first inner tooth, with a rise in the TiO2 concentration. The present study revealed most suitable endpoints in the case of TiO2 nanoparticle contamination in freshwaters, using Chironomus tentans as a biotidicator. The results show that morphological changes of C. tentans could be used as an endpoint in nano-TiO2 monitoring together with geometric morphometry.

WE408 Multigenomic expression of the nematode C. elegans to Silver Nanoparticles at the expense of oxidative stress defence mechanisms L. Rossbach, Norwegian University of Life Sciences UMB / INM; E. Maremonti, Norwegian University of Life Sciences UMB / MEF; M. Oughton, Norwegian University of Life Sciences; D. Oughton, Norwegian Public Roads Administration / Department of Biology and Ecology; B. Jovanovic, Iowa State University / Natural Resource Ecology and Management; A. Durđević, J. Stanković, Faculty of Science and Mathematics, University of Niš / Department of Biology and Ecology Adverse effects of Ag are widely known, with effects ranging from oxidative stress, immune suppression and inhibition of larval growth to neural defects, such as growth and reproduction. Most toxicological studies, however, only cover a limited timescale of the organism’s life stage, rather than the whole lifespan, or even across generation. Therefore, knowledge on multigenomic effects is lacking. The current study was conducted in order to determine whether the six generational exposure to sub-lethal concentrations of either ionic or nanoparticulate silver (AgNP) could induce alterations in sensitivity to Ag exposure using the nematode C. elegans as a model. Further, changes in susceptibility to other metals and the role of ROS as well as metabolic changes were investigated. Exposure to sub-lethal concentrations revealed increased susceptibility to ionic Ag, while...
AgNPs tolerance increased. Results show that adaptation development may occur after just a few generations. Subsequent exposure to paraquat, a known ROS inducer, indicated the involvement of ROS defense mechanisms. Therefore, changes in glutathione redox potential and sod-1 gene expression were measured, employing the genetically encoded fluorescent biosensors Grx1-roGFP2, and the reporter strain Sod-1::gfp, respectively. Further, effects of the AgNPs on the central metabolism and implications on energy production are investigated by metabolic footprinting of cells unexposed to AgNPs and treated with PE255. Findings of this study will aid to further improve the understanding of the toxicity of nanoparticles, as well as contribute to our knowledge about the behavior of E. coli in response to toxicants. Acknowledgements: Karl Andreas Jensen and Sofridh Lohné. This work was supported by the Norwegian Research Council funded NanoCharm (221398/E40) and NorNanoReg (239199) projects, and the EU NANOREG project grant agreement n° 310584.

**WE409**

Effect of silver nanoparticles layer on soil surface to terrestrial species

J. Kwak, S. Nam, Konkuk University; Y. An, Konkuk University / Department of Environmental Health Science

With developing nanotechnology, uses and release of engineered nanomaterials are increasing. Landfill of biosolid after wastewater treatment is considered as one of indirect pollution exposure sources of nanomaterials. This study focused on the simulation of exposure scenarios of nanomaterials landfills, and set the aim to investigate different toxic effects derived from different scenarios. Silver nanoparticles (AgNPs) were selected as test nanomaterials due to their different biokinetics; however, it was not revealed yet. Expanded polystyrene (EPS), one of common marine plastic debris, was known to weather more rapidly than polyethylene and polypropylene in our previous study. Fragmentation of nano- and micro-sized particles through weathering of plastic surface by photo-oxidation was regarded as one of major input sources of secondary nano- and microplastics. Its fragmentation process according to exposure duration and size of particles was investigated with different exposure for light and heat respectively. The mass of >1 µm sized particles was qualitatively measured by photo-oxidation. The mass of >10 µm sized particles produced per EPS cube surface area (g/m²) significantly (>0.05) increased according to exposure time: 0.1±0.1 g/m² for control, 2±0.3 g/m² for 2M, 3±0.4 g/m² for 5M and 7±0.2 g/m² for 9M. The mean and medium size of >10 µm EPS particles measured by laser diffraction was 26-29 µm and 18-20 µm, respectively. The hydrodynamic diameter of the EPS particles in the filtrates of <0.45 µm pore size filters were 0.32 nm for 2M, 530 nm for 5M and 752 nm for 9M by dynamic laser scattering. Their particle abundances measured by nanoparticle tracking analysis were 1.8±1.0 particles/ml for 2M and 3.2±1.0 particles/ml for 2M and 9M. Two month exposure of EPS to sunlight was enough to produce a large number of micro- and as micro-sized plastics by surface weathering.

**WE411**

Effects of nano-plastics on natural marine aggregates and their associated microbial communities

S. Summers, SCELSCE Nanyang Technological University / SCELSCE; T.B. Henry, Heriot-Watt University / The School of Energy, Geoscience, Infrastructure and Society; T. Gutierrez, Heriot Watt University / School of Life Sciences

Plastic debris in the marine environment is of particular interest, as the issue is one on a truly global scale. The ubiquitous presence of micrometer sized plastic particles and the knowledge that these will break down into ever smaller, nanometre sized particles, has resulted in a surge of recent research into nano-plastics. However, the difficulty in detection of nano-plastics has made it difficult to predict the specific risks involved in their presence within a marine environment. We recently showed that nano-plastics are readily incorporated into marine snow (MS) particles as mediated by ‘sticky’ polymeric substances and other particulates. MS particles are described as a heterogeneous matrix composed of polymeric substances, such as EPS, faecal pellets, invertebrate casts and microorganisms. It is therefore expected that the fragmentation of nano-plastics in MS would include plastics to the total pool of suspended particulates. This study examined the impacts of this pool of MS-associated nano-plastics, however, are lacking, including the microorganisms found colonizing these particles. Since microorganisms, in particular bacteria, are major colonizers of MS, we hypothesized this would also be the case for MS-associated nano-plastics particles. To assess this, we generated MS-associated nano-plastics particles collected from a subarctic northeast Atlantic region and nanometre-sized polystyrene spheres. Analysis of the nano-plastic-MS particles barcoded 16S rRNA gene MiSeq sequencing revealed that the addition of nano-plastics introduced some minor variability within treatments, with respect to microbial composition. The presence of the nano-plastics marginally increased the α-diversity of the community associated with the particles, compared to the community associated with MS in the absence of nano-plastics. Statistical analysis, however, did not provide substantive evidence to suggest that these differences and variabilities were significant. Therefore, while nano-plastics may have some minor effect in terms of the diversity of bacteria that colonize MS, it was less than the natural variability observed for the microbial communities that colonized non-exposed (no nano-plastics) MS particles. These results suggest that nano-plastics are not likely to have a major influence in altering the bacterial communities associated with MS particles.

**WE412**

Tracking nano-plastics in marine bivalves at environmentally realistic concentrations

M. AL SJD CHEIKH, University of Plymouth / Marine sciences and engineering; S.J. Rowland, University of Plymouth / School of Geography Earth and Environmental; K. Stevenson, Charles River; C. Roleau, Peches et Oceans Canada; T.B. Henry, Heriot-Watt University / The School of Energy, Geoscience, Infrastructure and Society; R.C. Thompson, Plymouth University / School of Marine Science and Engineering

Awareness campaigns on plastic pollution in oceans are backed by governments worldwide with recent initiatives to ban plastic products such as-micro-beads from cosmetics or single-use plastic bags. The fragmentation, the persistence and the production of plastic particles (micro <1mm, MPs, to nano-size < 1µm, NPs) are among the most prominent environmental issues faced by government environmental agencies. MPs are a vast group of contaminants and tracking of such small particles at these low concentrations are indeed major analytical challenges for environmental and laboratory studies. Our study focuses on the tissue distributions of nano-plastic in marine bivalve in a single pulse exposure of 6 hours and depuration of 48 days in open clean seawater. Here we present, preliminary results where we track nano-plastics at environmentally realistic concentrations in marine bivalves.

**WE413**

Plastics: does size matter? Impact of environmentally relevant nanoplastics identified in the Nordic environment

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Environmental stressors from plastics contribute to the plastic debris in the environment. These stressors are exposed to diverse plastic debris, which can be denominated as micro- (1-10 mm) or nano-plastics (<100 nm) depending on size range. Microplastics are ingested by a range of aquatic organisms and this ingestion might cause adverse biological effects, however less research has been conducted on their smaller counterparts, nanoparticles (NPLs). Similarly to other nanomaterials, NPLS possess size specific properties which could increase their toxic potential towards aquatic organisms depending on surface characteristics and interactions with the surrounding environment. Nonetheless, their presence in the environment and any toxic mechanisms are, to a large extent, unknown. In this study, the impact of environmentally relevant plastics identified in Norwegian plastics marginally increased the diversity of the community associated with the particles, compared to the community associated with MS in the absence of nano-plastics. Statistical analysis, however, did not provide substantive evidence to suggest that these differences and variabilities were significant. Therefore, while nano-plastics may have some minor effect in terms of the diversity of bacteria that colonize MS, it was less than the natural variability observed for the microbial communities that colonized non-exposed (no nano-plastics) MS particles. These results suggest that nano-plastics are not likely to have a major influence in altering the bacterial communities associated with MS particles.

Awareness campaigns on plastic pollution in oceans are backed by governments worldwide with recent initiatives to ban plastic products such as micro-beads from cosmetics or single-use plastic bags. The fragmentation, the persistence and the production of plastic particles (micro <1mm, MPs, to nano-size < 1µm, NPs) are among the most prominent environmental issues faced by government environmental agencies. MPs are a vast group of contaminants and tracking of such small particles at these low concentrations are indeed major analytical challenges for environmental and laboratory studies. Our study focuses on the tissue distributions of nano-plastic in marine bivalve in a single pulse exposure of 6 hours and depuration of 48 days in open clean seawater. Here we present, preliminary results where we track nano-plastics at environmentally realistic concentrations in marine bivalves.
environmental samples will be evaluated at the nanoscale in three key marine species, the cryptophyte algae *Rhodomonas sp.*, the harpacticoid copepod *Tisbe battalglia* and the blue mussel *Mytilus edulis* and compared to its microscale counterpart. The uptake, accumulation and elimination kinetics of NPLs in the three species will be evaluated under ecologically relevant conditions, as well as their potential translocation along the aquatic food chain. Furthermore, the acute and sublethal ecotoxicological effects of both plastic sizes will be investigated at individual, cellular and molecular levels using different biological endpoints. With the results obtained in this study we aim to discuss the differences in uptake, accumulation and biological responses between different sized plastics identified in the Nordic environment, and consequently bridge the current knowledge gap on the assessment of their potential hazardous effects in marine biota.

**WE414**
Ecotoxicity of engineered nanomaterials in relation to ecosystem complexity and functioning

W. Peijnenburg, RIVM / Center for Safety of Substances and Products; Y. Zhai, CML Leiden University; M.G. Vrijer, CML Leiden University / Conservation Biology.

Engineered nanomaterials (ENMs) are widely applied, and the release and accumulation of ENMs through waste effluent and deposition are threatening the aquatic and terrestrial ecosystem. While important knowledge has been gained about the uptake of ENMs on cultured invertebrate and microbial species based on simplified microcosm and mesocosm model systems, extrapolation of results of these experiments is often dependent on assumptions and differences between the intestinal and the plethora of environmental variables that affect the fate and toxicity of ENMs over various spatial and temporal scales. In this context, an overview is given on recent achievements in assessing nano-specific effects on systems varying in physico-chemical and biological complexity. Amongst others, it will be illustrated how ENM toxicity can be affected by the intrinsic physico-chemical characteristics of ENM (e.g. size, surface charge, coating) and extrinsic environmental characteristics (NOM, pH, electrolytes) and how ENMs interact with various components of food webs. ENMs in the environment may directly or indirectly affect a diverse array of organisms and microorganisms, which likely cascades towards distorted ecosystem processes. We further identify challenging yet promising research areas in this emerging field that are essential in pursuing a realistic risk assessment that accounts for ecosystem complexity and functioning.

The take home message is that there is a need of studies assessing not only impacts of ENMs on single species but also a need of a comprehensive framework of nano-specific toxicity in complex ecosystems. Considering the abiotic complexity of the transport of ENMs in the natural environment, studies performed with laboratory-cultured species need to include proper characterization and a proper quantification of the environmental factors that impact fate and effects of ENMs. In addition, the biotic complexity in the ecosystem especially in the aquatic and terrestrial environment indicates that risk assessment of ENMs should be conducted in an integral multi-dimensional perspective. For instance, relatively simple studies on uptake and accumulation of ENMs by invertebrate species need to further investigate the perturbation caused by interactions between the intestinal microbiome and the host. Also, the link between microbiorganisms and invertebrates in a detrital food web should be included for a systematically evaluation of ENMs toxicity.

**WE415**
Development of rapid reacting automatic mobile lab responding chemical accident of aquatic environment in Korea

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Most of available mobile lab developed and operated in Korea are air quality monitoring system and there is no rapid reacting mobile lab responding chemical accident of aquatic environment in Korea. We designed rapid reacting mobile lab with two major factors, 24hr operating and rapid starting within 1hr after arrival. We also considered system stability during transportation and accessibility to target river or stream, where we collected vibration information of a vehicle by exposure to road and raised spot of a road and reduced speed. Vibration vulnerability assessment were conducted with vibration testing shaker. We also collected topographical information related to diverse accessibility characteristic to river by conducting sampling at the selecting sampling point. Lastly, we adopted special air-conditioning system to control the system contamination from exposure to vaporized chemicals at the accident location. We adopted dual power supply system with 55kW, 24hrs operation capacity and 24hr operation capacity with 10kW supplementary battery system of 10kWh assisting and initiating the system on arrival and in between generator exchange. Vibration testing shaker are established with vibration information collected. Activated carbon proved to be most effective to control our target chemicals, which was composed onto COMBI type filter. These findings will be modulated and structured to maximize system stability.

[Keyword] chemical accident, mobile lab, rapid monitoring system.

**WE416**
Trophic Interactions in the Bioaccumulation and Depuration of Silver in Fish from a Lake Dosed with Nanosilver

C.D. Metcalfe, Trent University / Water Quality Centre; V.V. Yargeau, McGill University / Chemical Enineering; K. Newman, J.D. Martin, Trent University / Water Quality Centre

Bench-scale tests have shown that silver (Ag) accumulates in the tissues of fish exposed to silver nanoparticles (AgNPs). However, these experiments cannot replicate the complex biogeochemical processes and trophic interactions found in natural aquatic ecosystems. This study was conducted as part of whole lake addition project in which a total of 15 kg of AgNPs was added over two ice-free field seasons to a small lake (i.e. Lake 222) in the Experimental Lakes Area in Canada. Both yellow perch (*Perca flavescens*) and northern pike (*Esox lucius*) accumulated Ag in their tissues. The greatest bioaccumulation was observed in the liver tissues of pike, and a secondary peak in the gill tissues. A biotest was conducted with silver concentration observed in liver of 5.1 µg/g wet weight. In perch, the highest concentrations of Ag were observed in gill tissue. Monitoring in the lake using passive sampling devices and single particle ICP-MS confirmed that Ag nanoparticles were present in the water column and that Ag was distributed throughout the lake at estimated concentrations in the range of 1–11 µg/L. These data indicate that the primary mode of Ag bioaccumulation in perch was probably through uptake into the gill, whereas pike probably accumulated Ag from the diet. The transfer of Ag from forage fish to piscivorous fish can occur in natural lake ecosystems, leading to concentrations in some tissues that are 3 orders of magnitude greater than the concentrations in water.

**WE417**
Hepatotoxicity of iron oxide (magnhemite) nanoparticles in the guppy Poecilia reticulata

G. Qualhato, Federal University of Goias / Department of Morphology; T.L. Rocha, University of Algarve / CIMA; S.M. Saboia-Morais, Federal University of Goias / Department of Morphology

Although there are many applications of iron oxide nanoparticles (IONPs) in the nanomedicine and nanomedication, its ecotoxic effects to aquatic organisms remains unclear. In this study, the hepatotoxic effects induced by citrate-functionalized IONPs at environmentally relevant iron concentration (0.3 mg L⁻¹) was investigated in female guppy *Poecilia reticulata* by histopathological approach based on qualitative analysis and histopathological index after acute and long-term exposure. The animals were collected at the beginning of the experiment and after 3, 7, 14 and 21 days of exposure. TEM results demonstrate crystalline and rounded IONP with an average size of 3.97 ± 0.85 nm, and DLS and ELS analysis showed that the IONPs has low hydrodynamic diameter and high surface charge in ultrapure water (14.11 ± 0.2 nm, -51.1 ± 7 mV) compared to reconstituted water (21.4 ± 0.39 nm, -19.5 ± 6.5 mV). The histopathological results showed an increase in the occurrence of histopathological changes in fish after the 7 days of exposure to IONPs, such micro- and macro-steatosis, melanomacrophage aggregates, exudate and hemorrhagic foci. The acute (3 and 7 days) and long-term (14 and 21 days) exposure of *P. reticulata* to IONPs induced high histopathological indexes associated with circulatory disorders and inflammatory responses with high foci of melanomacrophages indicating an increase of hepatotoxicity according the exposure time. Furthermore, guppies exposed to IONPs showed increasing in the number of MMC when compared to the unexposed ones. This is a first study about hepatotoxicity of IONPs in guppies. The results indicated that the hepatotoxicity estimated by qualitative parameters and histopathological index are important biomarkers to indicate the animal health and the environmental impact of IONPs. The present study confirming that the guppy *P. reticulata* is a suitable model to test the hepatotoxicity of IONPs. Keywords: Nanomaterials; biomarkers; nanotoxicology; guppy. Session: Ecotoxicology and human toxicology: from molecules to organisms, from oms to in vivo (Fish model species in human and environmental toxicology) Presentation preference: Poster presentation

(eco)toxicity tests for hazard evaluation of recycling materials and waste (P)

**WE418**
Biotes for Hazardous Waste Classification (HP14): benchmarking Limits for Tolerable Ecotoxicity

R. Weltens, VITO / ABS; E. Rossi, OVAM; V. vanermen, VITO; K. Tirez, Flemish Institute for Technological Research VITO

The current HP14 classification is based on the chemical composition of the waste: i.e. the sum of the concentrations of individual chemicals with ecotoxic properties (substances labeled H400, H410, H411, H420) are compared to defined limit values (CLP). In the case of complex waste materials however the analytical data are not sufficient to make this classification. In this paper we report on the chemical evaluation as step 1 biotes in steps 2 (acute aquatic tests on eluate fractions) and step 3 (acute and sublethal tests on solid waste). Limit values are needed for tolerable ecotoxicity (TE) that are in line with the chemical limit values (step 1).

In the study presented here we benchmarked biotes results against waste materials that were proven to be toxic in step 1. In step 1, we used a suitable option for that material. The main conclusions were: the proposed set of biotes is essential for proper HP14 evaluation. Poor mass balance in the chemical data should be the trigger for the additional use of biotes. LID 4 is proposed as TE.
for steps 2 and 3 (to be further evaluated for a larger data set). Both aquatic and terrestrial tests are needed for complete HP14 evaluation. This study was funded by OVAM, the Flemish Waste Agency! The kind help of the technicians Guy Geukens, Cis Boonen, Wilfried Dumortier is highly appreciated.

WE419
What is the future for the waste wood in terms of ecotoxicological testing?

S. Legay, FCBA / Chemisty Ecotoxicology Lab; C. Martin, FCBA / Gironde
In Europe, the classification of waste is carried out by an assessment of the hazardousness of the waste using data of known waste composition according to the properties of danger. This classification can be based only on the waste composition if the available data are sufficient and relevant. This method is based on the sum of contributions from different classes with CLP (Classification, Labelling, Packaging) regulation [European regulation (EC) 1272/2008]. In the majority of cases under complex mixtures, or of unknown nature (e.g: exterior and interior joinery, furniture, panels, wooden paneling, wood flooring, construction waste and demolition….) including wood preservative, paints, glues, the characterisation of their wastes is considered to be difficult. Eco-toxicological testing seems to be the most relevant because the effects of all contaminants (synergistic effects, additive and antagonistic) are integrated. It is a major advantage in the characterisation of waste. In this case, the waste has to be then subjected to a battery of bio tests (aquatic and terrestrial) in order to evaluate one of the 15 existing properties: Eco-toxicity for the environment (HP14). Test strategies will allow wood wastes to be recovered or recycled.

WE420
QUALITY STANDARDS FOR URBAN WASTE FERTILIZERS: PUTTING ECOTOXICITY IN THE PICTURE

S. Chelinho, CFE / Centre for Functional Ecology / Department of Life Sciences of University of Coimbra; C. Venâncio, Department of Biology / Biology; J. Lopes, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro; J. Sousa, University of Coimbra / Department of Life Sciences
The recently adopted EU Circular Economy Package intends to boost the production and EU movement of fertilizing products, such as those obtained from urban wastes (UWF) and the harmonization of quality standards (certification) for such products, to avoid market rejection. The Portuguese and EU regulation on UWF production/commercialization relies only on physico-chemical/biochemical analysis, which do not give any insight on the fraction of contaminant/mixture of contaminants bioavailable for organisms, nor the existence of potential antagonistic and/or synergistic effects. The main objective of this study is to develop an environmental quality certification system for the use of UWF in agricultural systems. In the present contribution, it was intended to characterize the ecotoxicological potential of the selected UWF, by evaluating both soil habitat and retention functions using lower-tier laboratory tests. Five UWF, two with origin in source separated organic wastes (group I; theoretically with higher quality, ex. lower metal content) and three originated from the organic fraction of mixed municipal solid waste (group II) were selected and tested using a battery of standardized ecotoxicological assays with plants, soil invertebrates and freshwater species. Five soil-UWF dilutions (0.7; 2.1; 6.3; 18.9; 56.7%) and eluates of pure UWF were used as test-medium. The results show that the highest and lowest toxicity were observed in the two UWF from group I. Among soil organisms the range of sensitivities was: E. andrei > F. candida > E. crypticus > L. sativa > T. aestivum while for aquatic organisms was: H. viridissimus > R. subcapitata > C. vulgaris > H. incongruens > B. calyciflorus. The observed toxicity was probably related to UWF high salinity rather than with metal contents. The obtained data also reinforce the need to include information from biological susceptibility of the receptors potentially at risk on the available regulation to obtain a more realistic view of the potential risks and to adapt the UWF application practices. Ultimately, a sustainable economic growth based on the efficient use of resources/waste valorization can be promoted.

WE421
Chemical and Ecotoxicological Assessment of Reclaimed Asphalt for their Subsequent Use

V. Jares, M. Buckova, R. Lichinsky, J. Hegrova, J. Huzlik, K. EFFenberger, Transport Research Centre
Reconstruction and repair of the road infrastructure is a source of the reclaimed asphalt, which is suitable to continue to use. It is also necessary to deal with the environmental impact of these materials within their ongoing life cycle, except testing their mechanical properties. Currently, the environmental impact tests of reclaimed asphalt is done out in crushed condition, according to the leachability test of granular materials with grain size Scenedesmus subspicatus, Sinapis alba, Daphnia magna were carried out in aqueous extracts. The results were compared with the legislation and were evaluated in terms of the content of the monitored substances and the type of test material.

WE422
Leaching tests - a useful tool for the environmental impact assessment of construction products

Construction products and waste materials used for construction can be in contact with the environment and may release potential harmful compounds. Information on the total content of these substances in the product is not sufficient to assess its environmental impact since it does not consider realistic exposure conditions. Concerning the pathway to soil and groundwater by contact with rain or seepage water leaching tests are available. The aim of this presentation is to show exemplary results of existing leaching methods and underline the strength and weaknesses of this kind of test with selected examples of our work: Concrete roofing tiles with terbutryn were leached according to CEN/TS 16657-2 with permanent immersion into water and according to EN 16105 with nine immersion cycles each consisting of immersion and dry stages. The eluates were divided into subsamples for different parameters as pH, conductivity, total organic carbon, anions, cations and terbutryn. Concerning the assessment of this leaching data it is important to notice that the eluate concentrations do not represent necessarily environmental concentrations. Thus, the concentration in the leaching test cannot simply be correlated to limit values from environmental quality standards. Further considerations are necessary including exposure scenarios and environmental pathways before leaching tests can be used in risk assessment.

Advances in monitoring and evaluating remedy effectiveness for in situ amendments in soils and sediments

WE423
Assessment and management of stormwater on sediment recontamination:
you don't need to measure everything, just the right things

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Stormwater sources are difficult to understand because of the poor characterization of the irregular, event-driven inputs and the difficulty of managing diffuse sources of large volumes of runoff. The study objective is to develop methodologies to quantify the consequences of ongoing low-level sources on chemical concentrations, contaminant availability, and effects on biological receptors in surficial sediments. The study area was in Paleta Creek near Naval Base San Diego (NBSD), in California. Two storm-events were captured for particle size characterization and chemical analysis. Receiving and outfall waters collected using auto-samplers, which were triggered at each location during two different seasons. The samples were analyzed for a variety of metals, PAHs, and PCBs, as a function of particle size. Sediment traps and sediment cores were also collected from the Creek and subjected to bulk chemical analysis. The fractionated water and sediment samples were processed for metal extraction using the modified EPA method 3050A and 3050B, respectively, and were analyzed using ICP-MS and MERX-T. Persistent organic pollutants in water samples were Liquid-Liquid Extracted (LLE) using the modified EPA Method 3510C, while sediment was extracted by Pressurized Fluid Extraction (PFE with ASE 350) using the modified EPA Method 3545A. PAH analysis was performed on HPLC and PCBs on GC/MS. Results showed that storm-events were dominated by coarse particles initially most likely to lead to sediment recontamination in the near field of the receiving water (e.g., PAH, and Cd). Cu was associated to the dissolved and clay fraction, however the depositing loads were more influenced by resuspension and redistribution of sediment than water. Data suggested that PAHs and PCBs, due to low bioavailability as determined with passive sampling and bioaccumulation testing, are not a strong contributor to sediment toxicity which appeared to be better correlated to the presence of lipophilic compounds in the trap samples and indicated that the size-segregated contaminant loads and simultaneous receiving water measurements were very helpful in relating the stormwater discharges to sediment recontamination. The particle associations in stormwater along with spatial distribution particularly in sediment traps, and less in the sediment cores, can identify contributing locations, effective remedial approaches, and help to propose best practices for stormwater and sediment management.

WE424
Development of active capping materials for oil spill contaminated sediment remediation

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Petroleum is extensively used for making oil-based chemical and energy; its daily
consume is on average 20 million tons and this is not predicted to decrease. The increasing use of the petroleum is inevitably connected to an increase in oil spills. Oil spills can occur for many reasons including human error or equipment failure and whenever an oil spill occurs it can represent a worldwide environmental problem. Effective remediation strategies are required. The aim of this study is to develop different sorbent materials for the active capping of oil spill contaminated sea-sediment. The experimental investigation was performed on an oil spill contaminated sea-sediment. Different sorbent materials were tested for the active capping: a commercial Activated Carbon (AC) Carbonitalia, an organophilic clays (OC) CETCO Inc. and a biochar (BC). The sorption properties of the materials were first investigated in aqueous solution by performing equilibrium tests (isotherm) using a mixture of polycyclic aromatic hydrocarbons (PAHs) as target contaminants. The bioavailable concentration was assessed by using polyethylene (PE) (26 µm) passive samplers. Capping experiments were performed in glass cylinders, where a layer of contaminated sediment (7.5 cm) was capped with a layer of 3 cm of sand mixed with the different tested materials (AC, OC and BC) and synthetic sea water (4 cm) on top. The capping efficiency was assessed by using polydimethylsiloxane (PDMS) fibers as passive samplers. The PAH profiles in the different capping systems were compared in order to assess the capping efficiency. Biochar showed a greater capping efficiency than activated carbon and organoclay. For example, anthracene porewater concentrations in the first centimetre of capping material, were reduced by 69%, 56% and 99% respectively for activated carbon, organoclay and biochar after 1 month. The porewater concentrations were also used to model the long term (>12 months) behaviour of various cap configurations with a numerical simulator. These results give useful indicators that biochar can be considered a cost-effective alternative to the more widely used sorbent materials for capping oil spill contaminated sediments.

WE425 PCB Tissue Concentrations and Benthic Community Impacts at a Carbon Amendment Pilot Study in the Intertidal and Subtidal Zones of San Francisco Bay

C.J. McCarthy, CH2M / Environmental Services; C.A. Irvine, RBL / Ecosystem Services; T. Himmer, CH2M; s. clark, Pacific EcoRisk; R. Zajac, J. Eby, CH2M Historical site activities at the Hunters Point Naval Shipyard (HPNS) in South San Francisco Bay resulted in the release of chemicals, including polychlorinated biphenyls (PCBs), to offshore sediments. To inform remedy selection at this urban site, activated carbon (AC) amendments alternatives were evaluated in a pilot treatability study. Two 0.4 acre plots extending from the intertidal to the subtidal zone were treated with either AquaGate + PAC® or SediMit® were assessed for their potential to reduce ecological risks associated with PCB-contaminated sediment. Previous studies indicated that biodegradation could reduce the bioavailability of PCBs to the benthic clams (Macoma nasuta) in shallow intertidal sediments when aided by mechanical mixing. This study assessed the effectiveness of AC placements without mechanical mixing in deeper water that is more representative of conditions where full-scale remediation is expected. Tissue bioaccumulation, benthic invertebrate community composition, and chemical analyses were measured to determine indicators that biochar can be considered a cost-effective alternative to the more widely used sorbent materials for capping oil spill contaminated sediments. Comparisons were made between baseline, reference, and post-amendment conditions (8 and 14 months post-placement). PCB tissue concentrations in Macoma sp. were measured in situ (field) and ex situ (bench-top) after 28-day exposures. Developing field exposure chambers that allowed sediments to infiltrate the chambers and expose clams upon deployment and then removing the chambers and expose clams upon deployment was a challenge. Modifying a chamber design used in previous studies by Luthy et al. (2009) proved successful. Effective remediation strategies are required. The aim of this study is to develop different sorbent materials for the active capping of oil spill contaminated sea-sediment. The experimental investigation was performed on an oil spill contaminated sea-sediment. Different sorbent materials were tested for the active capping: a commercial Activated Carbon (AC) Carbonitalia, an organophilic clays (OC) CETCO Inc. and a biochar (BC). The sorption properties of the materials were first investigated in aqueous solution by performing equilibrium tests (isotherm) using a mixture of polycyclic aromatic hydrocarbons (PAHs) as target contaminants. The bioavailable concentration was assessed by using polyethylene (PE) (26 µm) passive samplers. Capping experiments were performed in glass cylinders, where a layer of contaminated sediment (7.5 cm) was capped with a layer of 3 cm of sand mixed with the different tested materials (AC, OC and BC) and synthetic sea water (4 cm) on top. The capping efficiency was assessed by using polydimethylsiloxane (PDMS) fibers as passive samplers. The PAH profiles in the different capping systems were compared in order to assess the capping efficiency. Biochar showed a greater capping efficiency than activated carbon and organoclay. For example, anthracene porewater concentrations in the first centimetre of capping material, were reduced by 69%, 56% and 99% respectively for activated carbon, organoclay and biochar after 1 month. The porewater concentrations were also used to model the long term (>12 months) behaviour of various cap configurations with a numerical simulator. These results give useful indicators that biochar can be considered a cost-effective alternative to the more widely used sorbent materials for capping oil spill contaminated sediments.

WE427 Remediation of mine wastes with biochar: effect on metal bioavailability to earthworms

M. Almira-Casellas, Leitat Technological Center / HEHS; V. Gonzalez, M. Diez-Ortiz, Leitat Technological Center

The impact of two biochar-based amendments (one from pruning trees and other from sewage sludge) on metal bioavailability to earthworms was investigated in aqueous solution by performing equilibrium tests (isotherm) using a mixture of polycyclic aromatic hydrocarbons (PAHs) as target contaminants. The bioavailable concentration was assessed by using polyethylene (PE) (26 µm) passive samplers. Capping experiments were performed in glass cylinders, where a layer of contaminated sediment (7.5 cm) was capped with a layer of 3 cm of sand mixed with the different tested materials (AC, OC and BC) and synthetic sea water (4 cm) on top. The capping efficiency was assessed by using polydimethylsiloxane (PDMS) fibers as passive samplers. The PAH profiles in the different capping systems were compared in order to assess the capping efficiency. Biochar showed a greater capping efficiency than activated carbon and organoclay. For example, anthracene porewater concentrations in the first centimetre of capping material, were reduced by 69%, 56% and 99% respectively for activated carbon, organoclay and biochar after 1 month. The porewater concentrations were also used to model the long term (>12 months) behaviour of various cap configurations with a numerical simulator. These results give useful indicators that biochar can be considered a cost-effective alternative to the more widely used sorbent materials for capping oil spill contaminated sediments.

WE428 Bioavailability-based Methods to Assess Remediation Effectiveness

J. Gan, University of California, Riverside / Department of Environmental Sciences; J. Wang, University of California Riverside; A.R. Taylor, University of California Riverside / Environmental Sciences; D. Schlenk, University of California-Riverside / Environmental Sciences

Compared to the total chemical concentration, bioavailability is a better measurement of risks of hydrophobic organic contaminants (HOCs) to biota in contaminated soil or sediment. Many different bioavailability estimation methods have been introduced to assess the effectiveness of remediation treatments. However, to date the different methods have rarely been evaluated against each other. Our findings suggested that both passive samplers and bioavailability methods may be used in assessing remediation efficiency, presenting flexibility in method selection. While accessibility-oriented methods offer better sensitivity and shorter sampling time, passive samplers may be more

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High performance biochar synthesized via co-serial foam, while biodegradation alone only reduced the recalcitrant fraction by Recalcitrant hydrocarbons were reduced by 92% using oxidation resulting in enhanced oxidation and biodegradation. Fractional analysis of TPH infiltration of hydrogen peroxide into the unsaturated soil. The easy and even bioaugmentation foam was sprayed every t contaminated soil for oxidation of soil total petroleum hydrocarbon (TPH). Periodic were applied to the surface soil by surfactant foam spraying. Surfactant foam would hydr...readily biodegraded in the natural soil system. Diesel includes recalcitrant... formation of PAH derivatives during SEE of PAHs. Polar PAH derivatives are produced from SEE, effluent collected during SEE, and pre...SEE soil. Mass balances were estimated for different PAHs, and some were above 100%, indicating the potential formation of PAH derivatives during SEE. These findings suggest that SEE pilot and treatability studies should include oxidants and surrogates to risk assessments to assess the full effectiveness of SEE and prevent underestimation of potential risks. A quantitative risk assessment will be performed by calculating B[a]Pα, concentrations and estimated lifetime cancer risk (ELCR) ingestion estimates. Developmental toxicity testing will be conducted with dechorionated zebrafish (Danio rerio) embryos (n=32/treatment) placed into 96-well plates containing pre- and post-SEE soil, effluent, and pre- and post-leaseate extract samples at 6 hours post fertilization. Developmental toxicity will be assessed by evaluating morphological changes, embryonic/larval photomotor behavior, and mortality at 24 and 120 hours post fertilization. This study quantified PAH derivatives after SEE, and it will identify implications for risk assessment and developmental toxicity outcomes.

Enhanced total petroleum hydrocarbon removal without soil disturbance by serial surfactant foam spraying R. Bajagain, Y. Jeong, Kunsan National University / Department of Environmental Engineering Fuel and air are a complex mixture. Polynuclear aromatic hydrocarbons are readily biodegraded in the natural soil system. Diesel includes recalcitrant hydrocarbons to the natural degradation and may act as potential and actual sources of harmful human and ecological effects. The purpose of the study was to evaluate serial surface foam spraying technology, which avoids disturbing the soil, to deliver chemical oxidant and oil-degrading microbes to unsaturated soil. Hydrogen peroxide was used as an oxidant and surfactant. All oxidants were applied to the surface soil by surfactant foam spraying. Surfactant foam would be a good media to spread remediation agents to the surface of contaminated sites with less labor or energy. Surfactant foam was sprayed once onto diesel contaminated soil for oxidation of soil total petroleum hydrocarbon (TPH). Periodic bioaugmentation foam was sprayed every 3 days for biodegradation of soil TPH. Foam spraying employing oxidation-bioaugmentation serial application significantly reduced soil TPH concentrations to 550 mg/kg from an initial 7470 mg/kg. Application of hydrogen peroxide by foam spraying increased the infiltration of hydrogen peroxide into the unsaturated soil. The easy and even infiltration of remediation reagents increased the contact with contaminants, resulting in enhanced oxidation and biodegradation. Fractional analysis of TPH showed C18-C22 present in diesel as biodegradation recalcitrant hydrocarbons. Recalcitrant hydrocarbons were reduced by 92% using oxidation-biodegradation serial foam, while biodegradation alone only reduced the recalcitrant fraction by 25%. (This work was supported by National Research Foundation of Korea (NRF-2015R1D1A1A01059664)).

Factors affecting sorption of halogenated phenols to polymer/biomass-derived biochar: Effect of pH, hydrophobicity, and deprotonation S. Oh, University of Ulsan / Department of Civil and Environmental Engineering; Y. Seo, University of Ulsan / Civil and Environmental Engineering; T. Sae, University of Ulsan / Department of Civil and Environmental Engineering. High performance biochar synthesized via co-pyrolysis of polymer and rice straw (RS) was evaluated as a sorbent for ionizable halogenated phenols. Compared with RS-derived biochar, the sorption of 2,4-dichlorophenol (DCP), 2,4-dibromophenol (DBP), and 2,4-difluorophenol (DFP) to polymer/RS-derived biochar was significantly enhanced by changing properties of biochar due to polymer residues, probably via hydrophobic sorption and electron donor-acceptor interactions. Removal of polymer residues and increasing aromaticity of polymer/RS-derived biochar at elevated pyrolysis temperatures affected the sorption capacity of halogenated phenols. Surface charge of biochar and deprotonation of the halogenated phenol species were other factors to be responsible for the sorption to polymer/RS-derived biochar. Competition with other halogenated phenols and dissolved cations implied that similar sorption mechanisms were existed and that surface complexation and electron donor-acceptor interactions were involved in the sorption to polymer/RS-derived biochar. Our results suggest that co-disposal of biomass and thermoplastic wastes through pyrolysis may be an effective option to produce a high-performance upgrade biochar as a sorbent for various types of contaminants.

Biochar for soil management: interactions with legacy contaminants and current-use pesticides L. Bielicki, RECETOX, Faculty of Science, Masaryk University / Faculty of Science, RECETOX; K. Kročová, Masaryk University / Faculty of Science, RECETOX; L. Skulcova, Masaryk University / Research Centre for Toxic Compounds in the Environment (RECETOX) Biochar (BC) is a product of thermal decomposition of biomass under oxygen-limited conditions. BC has received extensive attention because of its multifunctionality for agrochemical uptake by crops, improving disease resistance, its excellent sorption properties making BC a valuable sorbent in the treatment of solids contaminated with hydrophobic organic compounds (HOC) and wastewater. Due to its assumed stability, BC is considered an environmentally compatible approach for carbon sequestration and thus, climate change mitigation. Its application to agricultural soils has been shown to increase soil fertility, mainly due to improved physical properties, and its effectiveness is related to reduced water holding capacity. Despite these benefits, very little BC is currently utilized as soil amendment, mainly because the mechanisms improving soil health are poorly quantified and understood. Besides, there are several concerns and unknowns for BC agricultural application: i) the presence of pollutants (HOC, heavy metals) originating in pyrolysis or feedstock, ii) lower density resulting in transport of BC and BC-associated pollutants into surface water bodies, iii) substantial reduction of efficacy of agrochemicals; iv) effect of BC stability during weathering on its application potential, particularly long-term fate of agrochemicals. In this study, the role of BC in the management of contaminated soils and in the management of pesticide-treated agricultural soils is addressed. A systematic approach is devoted to the positive effects of biochar on both reduced availability of toxic contaminants and the ambiguous effects of reducing bioavailability of intentionally applied current-use pesticides. For that purpose, two different biochars were applied at increasing doses to soils amended with DDE and with epoxiconazole and tebuconazole as representative of legacy contaminants and broadly used fungicides, respectively. Bioavailability was assessed by means of solid-phase microextraction as well as functional activity for the uptake of chemicals by crop plants. This study aimed at balancing the dosing of biochar in soil management to ensure both an efficient and sustainable control of diffuse contamination and of pests.

PREPARATION AND CHARACTERIZATION OF COMPOSITES OF TYPE CLAY / POLYMERS AND THEIR USE IN THE REMOVAL OF CONTAMINANTS ORGANICS OF AQUATIC ENVIRONMENTS. M.S. Rodrigues, Instituto Federal do Maranhão; L. Aguilar Vieira, Universidade Federal do Maranhão; A. Costa Filho, S.G. RIBEIRO, Universidade Federal do Maranhão Clays have been used by mankind to many years due to its easy to obtain, its high technical viability rate and low economic cost. Among countless uses for the clay, research related to its various properties and also the adsorption capacity of this mineral, was greatly gained scientific attention. Mobility and diffusion and / or activation of these properties can be improved, resulting in a direct increase in the adsorption rate. The changes made in the samples of clays of this work were made from 2,6-bromoisoniofenon polymer, the process of cation exchange, which generated organophilic characteristics for samples, allowing comodosolventes of organic compounds were used. Serious damage to fauna and flora are caused by the use of organic phosphates of hydrocarbons, this leads to search for new materials aimed at removing it mainly aquatic environment. Given that the application of this work aims at the preparation and characterization of organophilic clays, as well as to evaluate its adsorption rate of these compounds organics PARA. It was used a natural clay from the Carolina-MA region. infrared techniques, X-ray diffraction, scanning electron microscopy (SEM), mass determination tests adsorbent and determining the concentration of Dimethylphosphate (DMP) as well a...
adoption capacity the material was 40% in nature, reaching a value of 78.4% after modification, deteriorating the feasibility of the process and material.

WE434

Field sampling and ex-situ bioassays for assessing the ecotoxicological risk of trace elements in different rehabilitated bauxite residues

E. Di Carlo, R. Courtney, University of Limerick / Department of Biological Sciences & The Bernal Institute; A. Boulemont, RíoTinto; L. Piotat, Alteo-Alumina

Bauxite residues are the by-product of the aluminium industry, resulting from the extraction of alumina from bauxite ore through the Bayer process. They could pose an environmental risk mainly because of their alkaline nature and the presence of trace elements (TE). For their rehabilitation, environmental regulations seem to be a promising, cost-effective and non-resource intensive option. However, there are still few studies evaluating the long-term success of the rehabilitation programmes and most of them do not consider ecotoxicology. The present study aims at assessing the success of rehabilitation strategies for bauxite residues considering the ecotoxicological risk of TE to organisms that live in the rehabilitated areas. To this end, two approaches were adopted: field sampling and ex-situ bioassays. On the one hand, samples of bauxite residues, plants and soil macroinvertebrates were collected from a range of field sites, over different seasons, and the environmental concentrations of TE were measured through ICP. On the other hand, exposure tests with earthworms (following OECD guidelines) and RHIZOTest (ISO 16198) with three herbaceous species were carried out, under controlled laboratory conditions, in order to investigate the bioavailability of TE. Data from the field represent the first step to understand whether there is a possible ecotoxicological issue for wildlife. Comparing the TE concentrations measured in the environment with the literature thresholds, some problematic elements (such as V) and sites (such as the one with compost capping) are identified. Regarding the laboratory bioassays, data show that the bioavailable fraction (taken up and accumulated by both plants and earthworms) does not always correlate with the TE concentrations measured in the extracts of the bauxite residues, indicating the inadequacy of chemical methods when comes to evaluate the ecotoxicological risk. In conclusion, the outcomes of this research are likely to shed light on the bioavailability of TE in a complex matrix, such as bauxite residues, and to provide a more realistic risk assessment for the organisms living there. Our data clearly show that the chemical total concentrations measured in the bauxite residues do not predict the bioavailable (potentially toxic) fraction of the TE, therefore bioassays should be taken into account when fixing the rehabilitation goals or assessing the rehabilitation success of a contaminated area.

Ecotoxicology of micro and nanoplastics: Mechanistic approaches to understand their risk for the environment and human health (P)

TH001

Synthetic textile fibers end up in agricultural soils - Can these microplastics pose a threat on soil organisms?

S. Colongon, University of Helsinki / Department of Ecological Science; C. Gestel, Vrije Universiteit Amsterdam / Ecological Science

An important route of microplastics (MPs) to the environment is the release of synthetic textile fibers to waste water due to laundry. The major part of the fibers is retained in the sewage sludge during waste water treatment and ends up in green spaces and agricultural fields. In this study, we explored the accumulation and effects of polyester fibers in soil involving exposure concentrations of 0.02 %, 0.06 %, 0.17 %, 0.5 % and 1.5 % of FES fibers in dry Lufa 2.2 soil. The fibers were mixed in with the soil to achieve as homogenous mixture as possible. Polyester fibers in soil affected the survival and reproduction of the entchytraeid worm Enchytraeus crypticus, but the effects did not show a consistent dose-related pattern. Compared to the control soil, the survival was decreased at fiber concentrations of 17 % and 9.5 %, whilst the reproduction was decreased in all other treatments except for the 0.06 % concentration. Polyester fibers did not affect the survival and reproduction of the springtail Folsomia candida and the oribatid mite Oppia nitens, the survival and feeding activity of the woodlice Porcellio scaber, nor the survival of the earthworm Eisenia andrei. Some of the measured endpoints seemed to show a negative response to the fibers, but the variation within the treatments was high, possibly also because it was difficult to achieve a completely homogenous distribution of the fibers in the test soil. The results of this study indicate that synthetic textile fibers can have slight negative effects on soil-dwelling entchytraeid worms, whilst arthropods were not markedly affected by the fibers. However, not only entchytraeid worms (E. crypticus), but also isopods (P. scaber), the springtail F. candida and the oribatid mite O. nitens, showed the presence of PE and PP particles in digestive glands of mussels exposed to the highest concentration tested (100 µg/L) of MPs, and in biodeposits where 112 500 particles/L of each MPs type. The exposure was followed by 10 days of depuration in clean seawater (without MPs). MPs fragments were prepared in the laboratory from commercially available products by milling; characterized in terms of content and several physiological parameters (metabolic activity, formation of intracellular reactive oxygen species and cytoplasmic membrane) in both photosynthetic organisms by flow cytometry using several fluorochromes. The results indicate that PHB released nanoparticles. NTA allowed to analyze the abiotic depolymerisation of PHB after 72 h in MilliQ finding a wide range (75 - 300 nm) of PHB nanoparticles. PHB induced a decrease in cellular growth and chlorophyll content in both photosynthetic organisms. Furthermore, PHB induced an increase in the level of intracellular reactive oxygen species and induced changes in membrane potential. In conclusion, microplastic particles of PHB exhibited toxicity towards photosynthetic aquatic organisms probably due to the release of a wide range of nanoparticles as a consequence of its own abiotic depolymerisation.

TH003

Differential responses of biomarkers in tissues of the blue mussel Mytilus edulis exposed to microplastics at Environmentally relevant concentrations

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Since the early 1970s, the occurrence of floating plastic has been reported in marine waters with great accumulation in gyres. In recent years, the presence of plastic debris <5mm called microplastics (MPs) which result mainly from macroplastic’s fragmentation has also been reported in aquatic ecosystems even in remote areas. Some studies have reported a presence of MPs and their effects in release of organisms. However, it appears necessary to investigate their potential toxicity especially at environmentally relevant concentrations. The aim of our study was to evaluate the bioaccumulation and toxicity of polypropylene (PP) and polyethylene (PE) fragments towards the blue mussel Mytilus edulis. These polymers were selected according to a previous study conducted in situ in the Region Pays de la Loire and Brest, where MPs were exposed in laboratory during 10 days at two environmentally relevant concentrations of 0.008 and 10 µg/L (9 and 11.250 particles/L, respectively) (Desforges et al., 2014), and to a higher concentration of 100 µg/L (112 500 particles/L) of each MPs type. The exposure was followed by 10 days of depuration in clean seawater (without MPs). MPs fragments were prepared in the laboratory from commercially available products by milling; characterized in terms of size, shapes and they were counted. Following exposure, tissues and biodeposits (faeces and pseudofaeces) were chemically digested and analyzed for MPs recovery using infrared micro-spectroscopy. Regarding potential toxic effects, detoxification and oxidative stress mechanisms through measurement of enzymatic activities of Glutathione-S-transferase (GST), Catalase (CAT) and superoxide dismutase (SOD) were investigated as well as the immune system and DNA damage responses. Results showed the presence of PE and PP particles in digestive glands of mussels exposed to the highest concentration tested (100 µg/L) of MPs, and in biodeposits where MPs were observed at all tested concentrations. Significant increases in SOD and CAT activities were observed in the digestive glands of mussel’s exposed to 0.008 and 10 µg/L and in gills from mussels exposed to 100 µg/L of MPs that could be indicative of an oxidative stress. This study brings new results on the potential sublethal effects of MPs at environmentally relevant concentrations of MPs.

TH004

Effects of zebrafish exposure to high-density polyethylene and poly(styrenes) microplastics at molecular and histological levels

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Effects of microplastic particles of polyhydroxybutyrate towards photosynthetic aquatic organisms

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Non-degradable, the ecotoxicological impact of microplastics in freshwater is not well understood [1]. Here we have investigated the effects of microplastic particles (nominal size 5 mm) of polyhydroxybutyrate (PHB) in two organisms representative of freshwaters, the filamentous cyanobacterium Anabaena sp. PCC7120 and the green alga Chlamydomonas reinhardtii. Firstly, we have performed the physicochemical characterization of microplastic particles of PHB and their potential degradation products in a MilliQ water by nanoparticle tracking analysis (NTA), dynamic light scattering (DLS) and infrared spectroscopy (IR). Then, we have evaluated the biological effects of PHB on cellular growth, pigment content and several physiological parameters (metabolic activity, formation of intracellular reactive oxygen species and cytoplasmic membrane) in both photosynthetic organisms by flow cytometry using several fluorochromes. The results indicate that PHB released nanoparticles. NTA allowed to analyze the abiotic depolymerisation of PHB after 72 h in MilliQ finding a wide range (75 - 300 nm) of PHB nanoparticles. PHB induced a decrease in cellular growth and chlorophyll content in both photosynthetic organisms. Furthermore, PHB induced an increase in the level of intracellular reactive oxygen species and induced changes in membrane potential. In conclusion, microplastic particles of PHB exhibited toxicity towards photosynthetic aquatic organisms probably due to the release of a wide range of nanoparticles as a consequence of its own abiotic depolymerisation.

Due to the constant increase of plastic use and production, microplastics (MPs) have become a contaminant of serious concern for the marine environment. However, detailed information about biological pathways affected by the exposure to different MP polymers is still lacking, in particular at transcriptome level. The present study focused on the identification of the molecular pathways affected by a chronic exposure of zebrafish (Danio rerio) to different concentrations of a combination of two environmentally relevant MPs for 20 days. Adult zebrafish were fed daily with dry fish food (control group, N=12) and food supplemented with a mix of pristine high-density polyethylene and polystyrene microplastics (two experimental groups: 0.1 and 1 mg/L, N=12 each). The microplastics dimension ranged from below 25 µm to 90 µm for both polymers. At the end of the exposure period, the liver was dissected and its whole transcriptome analyzed by next generation Illumina sequencing technology. In addition, the gastrointestinal tract and the gills were dissected and fixed for histology and immunohistochemistry. The exposure to polyethylene and polystyrene microplastics affected the liver transcriptome in a dose-dependent way, inducing the differential regulation of specific suites of genes. Histological analyses evidenced changes in the inflammatory response occurring at the two mucosal tissues selected for observation. The correlation of histological alterations with differential gene expression will be addressed and discussed. This study provides a comprehensive transcriptomic dataset useful for ecotoxicological studies on other fish species.

TH005 Uptake and Effects of Synthetic and Natural Microparticles in the Shrimp Palinonom varians
M. Weidung, University Duisburg-Essen; R. Saborowski, L. Gutow, Alfred Wegener Institute for Polar and Marine Research

Microplastics (< 5 mm) have become ubiquitous in waters. The smaller they are the easier they can be taken up by aquatic organisms. Once ingested they can cause various harmful effects. This study investigates the effects of size of artificial and natural particles on the induction of cellular stress in the Atlantic ditch shrimp (Palinonom varians). The study includes feeding experiments with different sizes of fluorescent microplastic particles, nanosized titanium dioxide particles and silica powder of diatoms as a reference for natural particles. The uptake and distribution of particles in the digestive organs was observed by fluorescent microscopy. As marker for oxidative stress we measured the activities of the antioxidant enzymes catalase and superoxide dismutase (SOD) in extracts of the midgut glands of animals which were fed with particles from 2 to 48 hours. The larger particles (2 µm and 10 µm) remained in the stomach and in the lumen of the gut. The smaller particles (0.1 µm) were translocated into the surrounding tissue and the epidermis of the gut gland. Decapods have a stomach with fine-meshed filter structures which prevent the uptake of particles > 170 nm into the digestive gland. Superoxide dismutase activity was rapidly induced when the animals were exposed to 0.1 µm plastic particles. The activity increased within 2 hours after microplastic ingestion and remained high after 48 hours. Slight difference appeared between natural and synthetic particles. The diatom powder also induced SOD activity which, however, continuously decreased with time. Due to the enzyme cascade where SOD reacts first, the activity of catalase was clearly lower. It can be assumed that any particles < 170 nm enter the cells of the midgut gland and induce oxidative stress. Following optical detection of reactive oxygen species (ROS) via confocal laser scanning microscopy will help to identify cellular reactions after exposure to microparticles.

TH006 Microplastics in the sub-surface layers of the South Atlantic Ocean
V. van der Schijff, North-West University / Unit for Environmental Sciences and Management; J. Karstens, GEOMAR - Helmholtz Centre for Ocean Research Kiel; R. Morard, Bremen University / MARUM - Centre for Marine Environmental Sciences; S. Speich, Ecole Normale Supérieure de Paris; H. Bouwman, North-West University / Unit for Environmental Science and Management

Microplastic particles in the ocean is a major environmental concern. Most studies tend to concentrate on the ocean surface when examining microplastic pollution. However, it is known that, for various reasons, microplastics can lose buoyancy and tend to concentrate on the ocean surface when examining microplastic pollution. This study investigated the effects of different sizes of plastic particles on the induction of cellular stress in the Atlantic ditch shrimp (Palinonom varians). The study includes feeding experiments with different sizes of fluorescent microplastic particles, nanosized titanium dioxide particles and silica powder of diatoms as a reference for natural particles. The uptake and distribution of particles in the digestive organs was observed by fluorescent microscopy. As marker for oxidative stress we measured the activities of the antioxidant enzymes catalase and superoxide dismutase (SOD) in extracts of the midgut glands of animals which were fed with particles from 2 to 48 hours. The larger particles (2 µm and 10 µm) remained in the stomach and in the lumen of the gut. The smaller particles (0.1 µm) were translocated into the surrounding tissue and the epidermis of the gut gland. Decapods have a stomach with fine-meshed filter structures which prevent the uptake of particles > 170 nm into the digestive gland. Superoxide dismutase activity was rapidly induced when the animals were exposed to 0.1 µm plastic particles. The activity increased within 2 hours after microplastic ingestion and remained high after 48 hours. Slight difference appeared between natural and synthetic particles. The diatom powder also induced SOD activity which, however, continuously decreased with time. Due to the enzyme cascade where SOD reacts first, the activity of catalase was clearly lower. It can be assumed that any particles < 170 nm enter the cells of the midgut gland and induce oxidative stress. Following optical detection of reactive oxygen species (ROS) via confocal laser scanning microscopy will help to identify cellular reactions after exposure to microparticles.

TH007 Effects of dietary microplastic exposure on fish intestinal physiology
G. Asmonait, H. Sundh, N. Asker, University of Gothenburg / Department of Biological and Environmental Sciences; B. Carney Almroth, University of Gothenburg Sweden / Department of Biology and Environmental Sciences

The accumulation of plastics in digestive tracts of aquatic biota has been extensively documented and ingestion has been proposed as a prominent exposure route for plastic debris for a variety of aquatic organisms. Large plastic items are known to physically block the intestinal passage, exert physical damage, impair food uptake and/or cause false satiation. Whereas, the biological effects resulting from ingestion of smaller micro-sized particles or microplastics (MPs) are considerably less documented and potential (negative) consequences on the alimentary tract are largely unknown. To address this, we designed a study aimed at assessing if/how ingestion of MPs can affect physiological function of the intestine in fish. We hypothesized that ingestion of MPs cause inflammatory responses and disturb intestinal barrier and transport functions. Juvenile rainbow trout (Oncorhynchus mykiss) were exposed via diet to polystyrene (PS) particles (50-250 µm, 10mg of PS MPs/fish/day) for a period of 4 weeks. Fish were fed four types of diets: control (no PS MPs) and diets containing untreated PS particles (PS Virginia) or particles exposed to UV treatment (PS-UV). Fish were fed the four diets at the end of the experiment. The intestines were dissected and fixed for histology. The findings of the study indicated no or minor functional effects on fish intestinal tissue inflicted by particle exposure. Signs of inflammation were detected and were accompanied by upregulation of tight junction proteins, suggesting activation of intestinal homeostasis in response to PS MPs exposure.

TH008 Biochemical responses and histological effects resulting from foodborne exposure to post-consumer microplastics in juvenile Solea senegalensis
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Microplastics (MPs) have dramatically increased in marine environments, being recognized as ubiquitous environmental pollutants. Two types of microplastics are usually detected in environmental samples: the primary MPs originally and intentionally manufactured MPs and the secondary MPs which result from the fragmentation of dumped plastic items. The latter are normally referred as post-consumer microplastics and usually present different shapes, colors, composition ad irregular surface. However, few toxicological studies have addressed exposure of marine organisms to post-consumer microplastics. The goal of the present research is to assess the effects of post-consumer microplastics in a model species (Solea senegalensis), using diet as the microplastic vehicle. For this purpose, four distinct diets were prepared using commercial fish pellets incorporated with two MPs sizes (< 200 µm and 300-500 µm) and two concentrations of each (562 and 56 MPs per day) and other without MPs (control), making a total of five treatments. Sixty-day laboratory assays were conducted, in duplicate, and the test pellets were provided to fish once a day. After 14, 30 and 60 days, fish were killed and samples from each treatment were excised. The liver and gills were fixed and stained with haematoxylin and eosin and were stored at -80°C for biochemical analysis. The digestive tract was also sampled and immersed in Davidson’s fixative for histopathological analysis. Biomarkers related with oxidative stress were analysed, namely the lipid peroxides, glutathione and the activity of catalase, superoxide dismutase and acetylcarnolinesterase. Histopathological analyses were also performed in the digestive tract to assess the presence of MPs and possible histopathological effects.

TH009 Nanoplastic impacts on physical, biochemical, and nutritional characteristics
of Pacific whiteleg shrimp

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Because of enormous amounts of plastic wastes in marine environment, the concerns about marine pollution and ecological damages on marine organisms have increased. Especially, among these plastic wastes, nanoplastics like microplastics (<5 mm) and nanoplastics (<100 nm) are getting a lot of attention. Many studies and the researches about their impacts and effects in environments are under way. In this study, we assess various physical, biochemical and nutritional changes in the bodies of Pacific whiteleg shrimp (Litopenaeus vannamei) exposed to nanoplastics. For 21 days, shrimps were fed mussels (Mytilus edulis) contaminated with nanoplastics (44 nm diameter) and their physical (length, weight, water contents, body mass index), biochemical (catalase, CAT; glutathione S-transferase, GST; superoxide dismutase, SOD; fecal microbiota viability), and nutritional (crude lipid, crude protein, amino acids, and fatty acids) changes were assessed after exposure. In results, nanoplastics attached on the filter and ingested to mussels entered the bodies of shrimps and affected the health and physicochemical properties of shrimps. Especially, biochemical changes were significantly induced in the bodies of shrimps. These results can be the evidence of the impacts of small sized plastics on marine organisms. This research was supported by Basic Science Research Program through the National Research Foundation of Korea (NRF) funded by the Ministry of Science, ICT, and Future planning (2016R1A2B1010445).

TH010

Brood Pouch-mediated Polyethylene Nanoparticle Accumulation During Daphnia magna Embryogenesis

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Polyethylene debris is ubiquitously distributed in aquatic environments and are considered as an emerging threat to aquatic organisms across trophic levels. While ingestion of particles receives most attention, other routes of uptake, such as respiratory uptake, warrants consideration of brood pouch-mediated accumulation. To study this, a well-established model organism in toxicity tests, Daphnia magna was used to track routes of uptake and target tissue of polyethylene nanoplastics (PSNP). A sub-lethal concentration of 5 mg L⁻¹ fluorescent PSNPs (25 nm) was used to monitor accumulation in adult animals as well as their embryos in the open brood pouch. A time series throughout embryonic development within the brood pouch revealed accumulation of PSNP in lipophilic cells in the early stages of embryonic development while the embryo is still surrounded by a chorion and before beginning of organogenesis. In contrast, PSNP particles were neither detected in the gut epithelium nor in lipid droplets in adults. An ex vivoexposure of embryos to PSNP showed similar changes in lipophilic cells, illustrating the likelihood of brood pouch-mediated PSNP uptake by embryos. Whether the observed brood pouch-mediated PSNP uptake ultimately translates to long-term effects under chronic exposure to environmentally relevant concentrations remains a challenging area for further research. By demonstrating embryo PSNP uptake via the brood pouch, data presented here give novel insights in bioaccumulation of nanoparticles and likely other lipophilic contaminants. Since this uptake route can occur within a diverse array of aquatic organisms, this study warrants consideration of brood pouch-mediated accumulation in efforts studying the hazards and risks of nanoparticle contamination.

TH011

Micro- and nanoplastic ingestion in blue mussel larvae

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A large number of aquatic species have been found to ingest microplastics in the field and in laboratory studies. Benthic invertebrates seem to be especially exposed to this form of pollution and the blue mussel Mytilus edulis is one of the species that has been investigated most in this respect. Studies have not only shown that the mussels ingest microplastics but have also reported diverse adverse effects on a cellular to a physiological level. However, the work has so far only focused on adult mussels and it is unclear how blue mussel larvae interact with and are affected by plastic particles according to their size and nanometre size range. Therefore, this research aimed at studying microplastic ingestion and potential physiological effects in blue mussel larvae. The first experiment aimed at quantifying the amount of ingested and egested particles. Ten day old larvae were exposed to two different sizes of fluorescent polystyrene microbeads (2 μm and 100 nm) and body burdens of particles were quantified after 4h. Subsequently, larvae were transferred to clean water and the amount of egested particles after 48h and 72h. The second experiment investigated potential effects of plastic particles on growth and development of the larvae. They were exposed to 3 different concentrations of the 2 μm and 100 nm beads, representing low (0.45 μg/L), medium (28.7 μg/L) and high (287 μg/L) exposure levels, for 2 weeks. Every 2-3 days larvae from the different treatments were sampled, fixed and photographed to analyse larval size and morphology. Results showed that the larvae readily ingested both particle sizes although ingestion appeared to be more efficient for the 2 μm beads. Egestion of micro- and nanoplastic particles did take place but was not complete within 72h, with 43% of the 2 μm and 61% of the 100 nm particles remaining in the animals. Potential effects on larval growth and development remain to be analysed. By taking other life stages into account and using a quantitative approach for analysing particle ingestion and egestion, this study contributes to enhancing the mechanistic understanding of microplastic – blue mussel interaction.

TH012

The sub-lethal impact of polystyrene microplastics and nanoplastics on the Mediterranean mussel M. galloprovincialis

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The contamination of marine environments by microscopic plastic debris is a current threat to the fitness of the exposed biota, and even higher concerns are risen on its potential fragmentation to the nanoscale. In the framework of the JPI OCEANs project PLASTOX, we investigated the chronic effects induced by a 21-day exposure to 1.5, 15 and 150 ng/L of polystyrene microplastics (MP, 3μm) and nanoplastics (NP, 50 nm) on the fitness of the mussel species Mytilus galloprovincialis. To do so, we employed a multibiomarker approach encompassing immunological responses (lysozyme and phagocytosis), lysosomal endpoints (lysosomal membrane stability and neutral lipids), oxidative stress (catalase activity, malondialdehyde and lipofuscin content) and detoxification (glutathione S-transferase) parameters and neurotoxic effects (acetylcholinesterase activity). The lysosomal membrane stability, whose impairment is a known general stress symptom, was generally reduced in mussel hemocytes after exposure to both MP and NP; however, only in MP-treated mussels this effect was accompanied by a decreased phagocytic activity. Lysozyme activity in hemolymph was affected by both MP or NP treatment. The general effect of nanoparticles on the immune system efficiency. All the performed treatments led to an accumulation of neutral lipids in the mussel digestive gland. Moreover, an accumulation of malondialdehyde and lipofuscin was observed at 150 ng/L NP and at 1.5 and 15 ng/L MP. In gill, catalase was up-regulated following either MP (1.5 and 15 ng/L) or NP (1.5 ng/L) treatments, while a decreased acetylcholinesterase activity was noted only at 15 ng/L NP. Biomarker data were integrated in the Mussel Expert System (MES), which estimates the stress level induced on mussels by calculating a A-E scale health status index (HSI). The MES did not identify health alterations in control and at 1.5 ng/L MP (HSI = A), while the onset of a low stress level (HSI = B) was detected at 15 and 150 ng/L MP. Differently, the stress level associated to NP (1.5 ng/L) was not significant. This study demonstrates that both polystyrene MP and NP induce a chronic stress syndrome in mussels by affecting lysosomal integrity and generating pro-oxidant conditions. However, the two particle types can differently alter immunological and neurological processes, with the exposure to NP resulting in a higher impact on the overall mussel fitness compared to MP.

TH013

Effect of cationic amino (PS-NH2) polystyrene nanoparticles in brine shrimp Artemia franciscana nauplii: biochemical and molecular responses

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The accumulation of plastic litter on beaches and open oceans has been identified as one of the major threats in marine ecosystems worldwide. Laboratory experiments have proved that the formation of nano-sized plastics during the polymer degradation may reach marine ecosystem, considered as the most in danger. In present study, the effect of 50 nm cationic amino polystyrene (PS-NH2) was investigated in nauplii of Artemia franciscana, which is commonly used as aquatic model organism in toxicity tests. Acute toxicity tests were performed on nauplii from sub-lethal suspensions of PS-NH2 (0.1, 1 and 10 μg/L) in natural sea water (NSW) for 48 hours. The toxicity was evaluated by measuring growth and several biomarkers as carboxysterolase (CHe), glutathione-S-transferase (GST), cholinesterase (Che), heat shock protein (HSP70), lipid peroxidation (LP) and catalase (CAT), involved in important physiological processes, such as biotransformation of xenobiotics, neuronal transmission and oxidative stress. The effects of PS-NH2 (at 0.1 and 1 μg/mL) on the expression of genes related to metabolism, biosynthesis and embryogenesis during the development of brine shrimp was also investigated. Genes included HSP26, HSP70, mitochondrial uncoupling protein 2 (UCP2), chaperon-containing TCP (TCP) and late embryogenesis abundant (LEA).

Acute exposure to sub-lethal suspensions PS-NH2 caused a significant decrease in growth in A. franciscana nauplii, as well as significant changes in all biomarkers studied, except for LP. A significant up-regulation of HSP26 and HSP70 was observed in nauplii exposed to 1 μg/mL of PS-NH2 as well as the modulation of TCP, the latter not significant. This supports the results obtained from biomarkers, suggesting a stress response and potential
apoptotic pathway following PS-NH₂ exposure. On the contrary, no significant effect on gene expression related to the brine shrimp’s metabolism (UCP2) was observed, and LEA was significantly modulated only at the lowest concentration tested. These findings indicate that stress-related responses are taking place in exposed nauplii after acute exposure to sub-lethal suspensions of PS-NH₂, and confirm the general concern about PS-NH₂ and their ability to represent an ecological treat for marine organisms. Given the increasing levels of plastic pollution in the oceans, additional studies should be done considering long-term exposure to analyze the potential risk of nano-sized plastics in marine environments.

TH014 The impact of nanoparticles on Antarctic krill Euphausia superba
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Under current climate change scenarios, Antarctic krill (Euphausia superba) is facing multiple stressors, which could affect its abundance and distribution. Microplastics have been recently reported in Antarctic waters, representing an additional potential impact on krill population. In this study we investigated the effects of model nanoparticles (< 1 µm) on krill juveniles through short-term exposure (48 h) of polystyrene nanoparticles (PS NPs) with different surface charge. The behaviour of anionic (60 nm PS-COOH) and cationic (50 nm PS-NH₂) NPs in Antarctic natural seawater (NSW, 34°S, 2°C) was also investigated by Dynamic Light Scattering. PS-COOH formed nanoscale aggregates (average size of 862 nm) in Antarctic NSF, while PS-NH₂ maintained their nominal size. No mortality was observed upon exposure to 2.5 µg/ml PS NPs after 48 h. However, krill exposed to PS-NH₂ showed lower motility than individuals exposed to PS-COOH and were characterised by significant up-regulation of β6 gene involved in new cuticle formation. Similar findings reported for other microcrustaceans have been associated with mortality over long-term exposure. Both PS NPs also accumulated in faecal pellets (FPs), which were characterised by lower density and sinking rate compared to control. Our findings demonstrate that PS NPs are able to affect swimming behaviour, cuticle formation and FPs properties of Antarctic krill, with potential serious consequences on Southern Ocean food web and biogeochemical cycle.

TH015 Exposure to nanoparticles as a potential stressor on Mytilus galloprovincialis
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Plastic pollution in marine environment is a worldwide problem, especially since plastic materials degrade into nano-size particles, becoming more bioavailable and constituting a source of entry of other contaminants into marine organisms. The present study aimed to assess the effects of polystyrene nanoparticles (PSNP) and clarify their modulation of short-term exposure. In both field and laboratory studies, a broad range of effects of model nanoparticles (< 1 µm) on M. galloprovincialis were observed. Only increased levels of the hepatic biomarker vitellogenin showed an effect on gene expression related to the brine shrimp's metabolism (UCP2) which was significantly modulated only at the lowest concentration tested. Laboratory tests and manipulative field experiments to study bioavailability of MP in the marine environment, tests materials with different sizes, shape (particles, fibres) and polymer compositions (polystyrene and polystyrene microbeads, polyster microfibres) were used. Using a novel approach, the influence of MP sorption on PAH bioavailability to two marine copepod species (Acartia tonsa and Calanus finmarchicus) was investigated using polystyrene particles with size ranges above and below the ingestion limit for the two species. The range of MP diameters used in the experiments was ~10-300 µm. Chemical body burden was measured after exposure to determine bioavailability. In conclusion, the presence of microplastics in marine environments has the potential to affect marine organisms and ecosystems, possibly leading to changes in their physiology, behaviour, and survival. Further studies are needed to fully understand the impact of microplastics on marine life and the overall health of marine ecosystems.

TH016 The role of microplastic size and type on PAH sorption and bioavailability to copepods
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It has been proposed that microplastic (MP) may act as a vector for a wide range of chemical pollutants already present in the environment. Polycyclic aromatic hydrocarbons (PAHs) are ubiquitous pollutants in the environment, known to cause adverse effects to a variety of marine organisms. Most PAHs have relatively high polymer-water partition coefficients, meaning their potential for sorption to, and transport by, MPs is high. In both field and laboratory studies, a broad range of marine species have been shown to ingest significant quantities of MP, with extended periods of retention observed in some cases. If PAHs are adsorbed to the MPs, this could present an alternative exposure route to PAHs for such species. However, the effect of MP sorption on PAH bioavailability in the marine environment remains poorly understood. Although several studies have attempted to study bioavailability of MP-adsorbed PAHs to aquatic organisms, most studies employ approaches that do not allow for accurate determination of if PAHs are released from adsorbed compounds or from compounds that have dissolved from the MPs into the exposure media. Here, we investigate the sorption kinetics and present adsorption isotherms for three model PAHs (fluoranthene, phenanthrene and 1,3-dimethylphenanthrene) to a range of different MP's in natural seawater. The selected PAHs exhibit different sizes and hydrophobicities, thus having varying scenario solubility (two to three orders of magnitude). In the case of fluoranthene, the mixed compound, fluoranthene, MP sorption could prove an important route of uptake in pelagic organisms. To account for the natural variability of MPs present in the marine environment, test materials with different sizes, shape (particles, fibres) and polymer compositions (polystyrene and polystyrene microbeads, polyster microfibres) were used. Using a novel approach, the influence of MP sorption on PAH bioavailability to two marine copepod species (Acartia tonsa and Calanus finmarchicus) was investigated using polystyrene particles with size ranges above and below the ingestion limit for the two species. The range of MP diameters used in the experiments was ~10-300 µm. Chemical body burden was measured after exposure to determine bioavailability. In conclusion, the presence of microplastics in marine environments has the potential to affect marine organisms and ecosystems, possibly leading to changes in their physiology, behaviour, and survival. Further studies are needed to fully understand the impact of microplastics on marine life and the overall health of marine ecosystems.

TH017 Limited influence of microplastics on the effects of an endocrine disruptor on the African clawed frog (Xenopus laevis)
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Endocrine disruptors (ED) are a class of chemicals that can affect juvenile sex determination and development of vertebrates like fish and amphibians but also humans. Tadpoles are not only vulnerable to ED, but potentially also to particulate contaminants like microplastics because they filter the surrounding water unspecifically during the first phase of their development. Until now, it is not clear if microplastics have negative impacts on amphibians and if they alter toxic effects of chemical contaminants like ED. This study focused on potential interactions between microplastics and endocrine disruptors (ED). Microplastics are ubiquitously distributed in freshwater ecosystems. Besides physical effects of the mere microplastic material itself, chemical effects by associated contaminants are discussed as potential threat to aquatic organisms, in particular after ingestion. Anthropogenic hormonal active substances, so-called endocrine disruptors (ED), can affect juvenile sex determination and development. In this study, we investigated the effects of model nanoparticles (PSNPs) on the African clawed frog (Xenopus laevis) and its ability to inhibit vitellogenin (Vtg). Vitellogenin is a biomarker for estrogenic compounds (EE2) and is known to be induced by endocrine disruptors (ED). We used model particles (EE2) with a size range of 15-20 µm (mean diameter) with an irregular shape were used as model MP. Tadpoles were exposed in batches with chronic exposure for 21 days to one concentration of EE2 (10^5 M) and a low and a high concentration of PAH-particles (1 and 100 mg L^-1) separately and in combination with each other. Stress hormones and larval development as well as sexual differentiation were assessed by morpho-grossmorphology and histology. Biomarkers, e.g. vitellogenin, were analysed as well. The concentration of EE2 in water was assessed by serum analysis. EE2 physical effects of the microplastic particles themselves on larval development and sexual differentiation were not observed. Only increased levels of the hepatic biomarker vitellogenin showed higher exposure of EE2 in treatments including PA particles in comparison to treatments without microplastics. All other EE2 specific endpoints were not influenced by PA particles. These results indicate that microplastics only play a minor role for the effects of a hormonal active chemical in amphibians and thus provide insights for an indepth risk assessment of MP in the environment.
Arctic), a range of different virgin polymer pellets (LDPE, PP, PS and PET), as well as marine litter-derived microplastic particles from the fish box, were deployed underwater in the small boat harbor at Tromsø, Northern Norway for up to 12 months. The deployment device consisted of an empty stainless steel SMFD canister, with the various plastic types placed in reusable, empty teabags made of PP, placed separately in nylon netting. Sampling was conducted 1 week, 1 month, 3 months, 9 months and 12 months after deployment. Hydrophobic persistent organic pollutants such as PAHs, PCBs, DDTs, PBDEs and organochlorines were extracted and analyzed to establish the sorption kinetics in seawater under Arctic conditions. Samples were extracted using ultrasound and nonpolar solvents, followed by GPC and SPE clean up. Analytical chemistry using GC/MS/MS and GC/MS was done in the laboratories of the TU Darmstadt and NILU, Tromsø. In addition, release kinetics of common phthalate esters, organochlorine pesticides, bisphenol A and perchlorinated by-products were estimated from other four post-industrial virgin pellets (LDPE, PS, PVC, PET) according to the same sampling protocol. Chemical analysis was performed using either GC/MS or LC-QTOF.

TH019
Characterization of microplastics present in personal care products and the study of its toxicity mixed with chlorpyrifos on juveniles of Solea senegalensis. G. Albení, Spain; I. Cabrera Pozo, University of Cadiz; D. Coello, R. Rodríguez-Barroso, J. Quiroga, University of Cadiz / Environmental Technology; J. Arellano, University of Cadiz / Toxicology Area.

In the last decade, different components from personal care products have arrived at aquatic ecosystems because these products are not biodegradable or removed in wastewater treatment plants. Some of the personal care products contain plastic microbeads such as exfoliating shower gel, toothpaste and make-up cream commonly used and available in supermarkets of our area were used in these assays. The microspheres available in these samples were separated and chlorified. The particles were identified by Fourier transform-infrared spectroscopy (FT-IR) using a PerkinElmer Spectrum 100. The spectra are recorded in reflection mode in the spectral range 4000-650 cm⁻¹ by co-adding 128 scans at a resolution of 4 cm⁻¹. The particles were identified by comparing FT-IR absorbance spectra of the microplastics to those in a polymer reference library. The microplastics were used in toxicity tests. In aquatic ecosystems and their organisms are exposed to complex mixtures of environmental contaminants as pesticides and microplastics. Thus, the effects of microplastics interaction with chlorpyrifos, an organophosphate pesticide, have been studied in this work. The toxicity studies were carried out during 96 hours with continuous ventilation and water renewal every 24 hours, at a temperature of 19-20°C and under 12h light/12h dark exposure. The juveniles of Solea senegalensis (weight 3.07 ± 0.49 g) were exposed to five nominal concentrations of chlorpyrifos (5–80 µg/l), three concentrations on this compound mixed with microplastics (chlorpyrifos: 5-1020 µg/l; microplastics: 0.150 mg/l, microplastics alone (0.150 mg/l), plus an untreated control and a solvent control (acetone). In these assays not mortality was observed on juveniles with both compounds and their mixtures. Cholinesterases (ChE) have been used as specific and sensitive biomarkers of exposure to organophosphates pesticides. In general, there are two type of ChE presented in fish, acetylcholinesterase (AChE) and butyrylcholinesterase (BChE). The AChE was analyzed on the head homogenate of juveniles, after this crude was inhibited with iso-OMPA, which is a specific inhibitor of BChE. The results showed that there was inhibition activity in the head of Solea senegalensis in presence of chlorpyrifos. However, it was not observed significant differences between the same concentrations of chlorpyrifos and its mixture with microplastics.

TH020
Are microplastics inhibitory to Daphnia magna and are they significant vectors for hydrophobic organic pollutants? C. F. Frydkjær, Aalborg University / Biology and Environmental Science, Denmark; C. K. Frydkjær, Aalborg University / Biology and Environmental Science; N. Iversen, Aalborg University / Department of Chemistry and Bioscience; P. Roslev, Aalborg University / Biology and Environmental Science.

The presence of microplastics in aquatic ecosystems is of increasing global concern. Ingestion of microplastics may result in adverse effects in aquatic organisms, and the increasing number of studies aiming at quantifying microplastic uptake and assessing the effects resulting from dietary exposures to PS particles ranging in size 50-250 µm. In this study, Chironomus tentans (Onconochyhus mykiss) were exposed diets, enriched with PS particles (10mg of PS particles/Fish/day) for 28 days. We used environmentally contaminated PS particles from in situ exposures from two environmental matrices (undiluted sewage effluent and industrial harbor runoff). As PS particles largely exceed sizes relevant for biological uptake, it provides an opportunity to study particles’ role as vector for environmental pollutants. Three different experimental diets, containing PS, sewage (PS-sewage) and harbor (PS-harbor) exposed particles, were developed. Thereafter, a suite of oxidative stress biomarkers was investigated in the hepatic tissue of fish exposed to PS particles vs gene expression analysis (NRF2, GR, GST, GS, GPx, CAT, GCLmd, GCLcat, SOD) and enzymatic assays (GR, GST, GS, GPx, CAT). Additionally, mRNA levels of established biomarkers (CYP1a, ERα and β, AR, MT, VTG) were quantified to provide additional insights into xenobiotic-related hepatic responses to dietary PS particles exposures. The findings of this study revealed an indication of NRF2-mediated oxidative stress regulation.
Fish from PS-sewage and PS-harbor treatments had altered expression levels of multiple antioxidant enzymes in liver. Dietary exposure to PS MPs resulted in low-level activation of hepatic oxidative stress, which may not necessarily exert harmful effects on hepatic physiology, but may rather indicate adaptive homeostatic regulation. Differential responses to different PS MP treatments (PS-sewage and PS-harbor) potentially could be explained by different chemicals associated with particles during in situ exposures.

**TH023**
Effects of Nanopolystyrene and the Co-Contaminant Tributyltin on the Nematode Community Structure in Sandy Sediments
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Nanoplastics (MPs, ≤ 1 µm) can result from larger plastic debris released in the environment and can pose a risk to marine organisms and ecosystems. The risk of NPs can be exacerbated because toxicants sorbed to NPs may be transported to and become more bioavailable in organisms. It is likely that NPs are the most abundant plastic particles present in marine environments, and as in the case of microplastics, they are expected to accumulate in benthic ecosystems. However, there is no information on the impact of NPs on benthic meiofauna assemblages. It is critical to understand impacts of NPs on sediments of NPs because meiofauna communities play key roles on ecosystem functions such as food production and nutrient cycling. Nematodes are well established as pollution indicators and structural shifts in their communities reflect environmental changes. The goal of our work was to assess the effects of nanoplastyrene (nPS) and nPS with the sorbed co-contaminant Tributytin (TBT) on free living nematodes on sandy sediment within a mesocosm experiment. Sediment was collected (up to 5 cm depth) at the Eden Estuary, St Andrews, Scotland, UK. The mesocosms (12°C) consisted of glass beakers (1 L) and the exposure took place for up to 2 months. Core samples of sediments were taken each week from the following treatments: 1) Control sediment, 2) Sediment with spiked TBT (0-100 ng/kg), 3) Sediment with nPS (0-12 mg/kg) and 4) Sediment with nPS spiked with TBT (0-10 mg/kg nPS). Oxygen penetration depth (OPD) was determined by measurement of the oxygen saturation in the sediments using a microprofiler equipped with oxygen microsensors. Changes in the nematode community structure were measured by assessment of changes nematode diversity (nematodes identified to genus) and dose response analyses according to nPS and/or TBT concentrations in the sediments. We anticipate that our results (ongoing data analysis) will contribute to a better understanding of the environmental risk of NPs and their co-contaminants within a relevant scenario.

**TH024**
Nanopolystyrene Induces a Decrease in the Oxygen Uptake of Zebrafish Larvae and Enables Sorbed Benzo(a)Pyrene Bioavailability
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Microplastics (5 mm–1 µm) and nanoplastics (NPs, ≤ 1 µm) can result from larger plastic debris released in the environment and can pose a risk to aquatic organisms. Potential effects of MPs include disruption of gut physiology after ingestion, release of substances (co-contaminants) sorbed to MPs into organisms, and occlusion of tissue surfaces by accumulation of MPs. Although not yet effectively measured in aquatic environments, NPs may be the most abundant plastic particles present, but little is known about their effects in organisms. Because the relative surface area is greater for NP than MPs, there is greater potential for co-contaminant sorption to NPs and subsequent co-contaminant release into organisms upon ingestion. We evaluated the bioavailability of the co-contaminant Benzo(a)Pyrene [BaP] sorbed to nanoplastyrene (nPS, 500 nm) by measuring the contributions in the sediments. We anticipate that our results (ongoing data analysis) will contribute to a better understanding of the environmental risk of NPs and their co-contaminants within a relevant scenario.

**TH025**
Impacts of Exposure to Microplastics alone and with adsorbed benzo[a]pyrene on biomarkers and scope for growth in marine mussels M. galloprovincialis\* J. Hatfield, N. González-Soto, University of the Basque country UPV/EHU; A. Katsumi, University of the Basque Country / CBET Research Group, Dept. Zoology and Animal Cell Biology Faculty of Science and Technology and Research Centre for Experimental Marine Biology and Biotechnology PIE; E. Duroudier, University of the Basque Country (UPV/EHU); J. Lacave, University of the Basque country UPV/EHU; A. Orbea, University of the Basque Country UPV/EHU / CBET Research Group, Dept. Zoology and Animal Cell Biology Faculty of Science and Technology and Research Centre for Experimental Marine Biology and Biotechnology PIE.

Due to their hydrophobicity and relatively large surface area, microplastics (MPs) can act as carriers of hydrophobic pollutants in the ocean and may facilitate their transfer to organisms (so-called “Trojan-horse effect”). This study examined the effects of different levels of biological O2 uptake (0.5 and 4 µM), and the exposure took place for up to 2 months. Core samples of sediments were taken each week from the following treatments: 1) Control sediment, 2) Sediment spiked with BaP solutions (45 μg/ml with sorbed BaP), whereas this gene did not express when larvae were exposed just to nPS, indicating desorption of BaP. We anticipate that our results (ongoing data analysis) will contribute to a better understanding of the effects and risk of NPs and their co-contaminants within a relevant scenario.

**TH026**
Characterization of the adsorption/desorption of benzo[a]pyrene from polystyrene micro- and nanoplastics for further toxicity assessment
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Degradation processes that large plastic items undergo in the sea have led to the appearance of small plastic pieces known as micro- (MPs) or nanoplastics (NPs), depending on their size. MPs and NPs can also be specifically manufactured for industrial and domestic applications, which results in an additional source of pollution. MPs and NPs become available to biota and enter into the food web. Microplastics and their products can be transported over large distances by hydrodynamic processes and they can adsorb/absorb other pollutants present in the water column acting as Trojan Horses. In order to further investigate the ecotoxicological aspects of this phenomenon, the characterization of the adsorption of benzo[a]pyrene (BaP), as a model polycarboxylic hydrocarbon, to polystyrene MPs and NPs (4, 0, 5 and 0.05 µm), was undertaken. 50 mg/l of particles of the three sizes were incubated for 24 h in an orbital shaker at 300 rpm (21°C) in three BaP solutions (100, 10 and 1 µg/l containing 0.01% DMSO) in MilliQ water. After the adsorption period, centrifugation was performed in order to settle the plastic and allow the removal of non adsorbed BaP (NA-BaP), NA-BaP was quantified in the...
supernatant by SPME/GC/MS. To measure BaP adsorbed to the plastics (A-BaP), plastic solutions were freeze-dried and subjected to microwave-assisted extraction before GC/MS analysis. The ad/absorption capacity of the plastics was calculated in mass of adsorbed BaP per gram of plastic (µg g⁻¹) for the different sizes of plastic in order to determine the capacity of ad/absorption of polystyrene microbeads and whether this process was directly dependent on plastic size. Results indicated that the sorption of BaP as A-BaP on a high-density polyethylene (HDPE) surface was > 4.5 µm MPs. The percentages of A-BaP vs total BaP solution were 90.88% and 37.18% with a Qmax of 217.39 µg g⁻¹ and 18.33 µg g⁻¹ (Langmuir model; R²=0.9862, 0.9477) for 0.5 µm and 4.5 µm MPs, respectively. In both cases the applied methodology was successful to characterise the ad/absorption process of BaP to MPs and is currently being applied to NPs. * Funded by French ANR (No. 10-EX-03-02 and Cluster Excellence COTE (ANR-10-LABX 45), Spanish MINECO (NACE project — CTM2016-8130-R), Basque Government (consolidated research group IT810-13) and UPV/EHU (UI11/37 and grant to IMa). \[TH027\]

Occurrence of microplastics in epibenthic and sediment-dwelling species in a Norwegian fjord a. bour, ECOLAB UMR254 CNRS UPS INPT; C.G. Avio, L. Pittura, S. Gobri, Department of Life and Environmental Sciences, Polytechnic University of Marche, Ancona, Italy; F. Regoli, Università Politecnica delle Marche; K. Hylland, Department of Biosciences, University of Oslo, Oslo, Norway / Department of Biosciences

The exponential production and use of plastics has generated millions of tons of plastic waste over the past decades, and the presence of microplastics has been reported throughout the world’s oceans. The ingestion of microplastics in situ has been shown in various species, but important knowledge gaps remain, as most studies focus on pelagic fish species or bivalves used for human consumption. Here, we report the presence of microplastics in ten sediment-dwelling and epibenthic species representative of different feeding modes and trophic levels. The species analyzed include fish, bivalves, echinoderms, crustaceans and polychaetes. Organisms were sampled in the inner Oslofjord (Oslo, Norway), which is a fjord subject to strong anthropogenic pressures. High occurrence of plastic contamination was observed, with microplastic particles found in all species and in half of the individuals on average, and present in 75% of the individuals for some species. The extracted microplastics had various shapes (fibers, fragments, flakes), colors and sizes. Micro-FT-IR analysis revealed the presence of various plastic polymers: polyethylene, polypropylene and polyamide were the most commonly found, with 37%, 25% and 15% respectively. We hypothesize that marine and fish species have the main source of release for these compounds. Indeed, ropes and fishing lines are usually made of these polymers. Six other types of microplastics were also found, less frequently: PET, PBT, EVA, polystyrene, polyacrylic, and copolymers. These results underline the potential risk posed by microplastics in sediments and the importance of assessing microplastic occurrence and impacts in benthic environments.

\[TH028\]

Development of an optimal analytical protocol for the extraction of persistent organic pollutants adsorbed on plastic debris in the environment K. Sakaguchi-Soeder, Technische Universitaet Darmstadt / Institute IWAR Chair of Material Flow Management and Resource Economy Germany; T. Neveugl, Technische Universitaet Darmstadt / Department of Civil and Environmental Engineering; A. Goharjua, Technische Universitaet Darmstadt / Institute IWAR Chair of Material Flow Management and Resource Economy; M. Gottschling, Technische Universitaet Darmstadt / Institute IWAR Chair of Material Flow Management and Resource Economy Germany; A. van Oyen, Plastic Partner GmbH

Accumulation of persistent organic pollutants (POPs)-loaded microplastics (MPs) in the aquatic milieu is an emerging issue of international concern. Qualitative/quantitative determination of POPs on MPs is essential for the qualitative/quantitative determination of POPs on MPs is essential for the aquatic milieu is an emerging issue of international concern. Determination of sorption capacity of persistent organic pollutants (POPs) on microplastics (MPs) is essential to study ecological effect of POPs-loaded MPs in the aqueous environment. However, due to high octanol-water partition coefficients (Kow) as well as low water solubility, determination of sorption capacity of POPs on MPs in the laboratory is challenging. Here we present two methods to determine plastic-water partition coefficient of three polycyclic aromatic hydrocarbons (PAHs) on low-density polyethylene (LDPE): conventional spiking method and cellulose dialysis tubing method in batch test in the laboratory. PAHs selected for this test were naphthalene (log Kow = 3.37), phenanthrene (log Kow = 4.46) and fluoranthene (log Kow = 5.16). The plastic samples tested here are LDPE pellets containing given amount of LDPE and MilliQ water were prepared. A high concentration of single PAHs in ethanol solution was injected into each batch reactor until the PAH concentration became stable. PAH concentration in the batch was controlled using high performance liquid chromatography (HPLC). For the dialysis tubing method, the other hand, a closed dialysis tubing (permeability of 12,000-14,000 Dalton) containing a given amount of LDPE and MilliQ water was placed in each batch reactor filled with MilliQ water with single PAHs far above solubility. The water concentration outside of the tubing was expected to stay constant (= water solubility) during the entire experiment. The PAH concentration in the dialysis tubing was controlled using HPLC until the PAH concentration became stable. All batch reactors were placed on a horizontal shaker. When the adsorption of PAHs is completed, PAHs on LDPE are to be extracted and quantified using gas chromatography-mass spectrometry (GC/MS). Sorption capacity of each PAH was derived from the experiments and methods were compared. PE-water partition coefficient of these PAHs were derived based on the sorption capacity using adsorption models. TU Darmstadt and CARAT are participants of an EU project „PLASTOX“, a consortium of a JPI Ocean’s Joint Action.

\[TH030\]

Microplastics in food and beverages - a distorted perspective on risk S. Rost, DTU (Technical University of Denmark) / Department of Environmental Engineering; B.C. Alnroth, University of Gothenburg / Department of Biological and Environmental Engineering; E. Einarsson, DTU (Technical University of Denmark) (DTU) / DTU Environment; C. Van Texten; M.T. Carlsson, University of Gothenburg

Microplastics are ubiquitous in aquatic environments and they are ingested by a wide range of animals, including species for human consumption, i.e. bivalves and fish. Additionally, plastic particles have been reported in other food products and beverages, like honey, salt, beer and drinking water. This has triggered a discussion on the human health implications of this contamination – an aspect that has gained increasing attention in the scientific and public debate in recent years. The focus and extent of this debate, however, stands in contrast with scientific findings, which merely show the presence of microplastics in certain products but no actual effects on humans. It is without question that plastics can constitute a health risk but the scientific community has yet to participate in toxicological studies to determine the degree to which microplastic exposure via food products and beverages contributes to this health risk is likely insignificant at present time. When considering the magnitude of plastic usage and consequential exposure to plastic materials in our everyday lives, the relatively few microplastic particles that have been reported in food products and beverages will likely only constitute a minor exposure pathway for microplastics and associated chemicals. But as this is rarely put into perspective, the current debate creates a distorted picture of plastic exposure and risks to humans, resulting in a misdirected outrage when people find out about plastic particles in fish, while they at the same time do not reflect on the plastic container in which the fish is packaged for transport to stores and homes. In this way, the focus is taken away from the root of the problem, namely our use, consumption and disposal of plastic materials. We therefore want to encourage a more balanced and careful discussion on human health implications of plastics that takes these aspects into account.
Is the Arctic threatened by plastics? Identifying sources and determining the distribution of microplastics around Svalbard

Where is all the plastic, and what concentration of microplastics are ecosystems being exposed to? These are puzzling questions to the scientific community when comparing estimated values on annual plastic pollution with the actual measurements in the world’s ocean habitats. Recent investigations find plastic far away from any known sources, such as in deep ocean sediments and buried within polar sea ice. The Arctic Ocean has, despite its remoteness, been suggested as a sink for plastic. Fed by the thermohaline circulation driven deep-water formation, the Arctic Ocean’s bottom might be a dead end for plastics. However, very few studies have quantified the actual environmental concentrations of plastics in this remote area, thus the exposure to organisms living in this environment and potential effects are unknown. In this study, we investigated the occurrence, potential sources and distribution of microplastics in the Norwegian Arctic. We quantified and characterized anthropogenic particles >10μm in different environmental compartments (sediment, water, benthic invertebrates and sea ice) around the western and northern coasts of Svalbard. Samples were collected close to a sewage outlet and far from human activities close to the sea ice front in the Arctic Ocean. By sampling at several distances associated with different water masses (Atlantic, Arctic and sewage water as well as sea ice) could be quantified. Simultaneous measurements of organic matter tracers for sea ice microalgae (IP25), pelagic microalgae (C25:3) and sewage (coprostanol) enabled correlations to be made on potential sources, pathways and fate of microplastics in the Arctic. Additional analyses of the presence of plastic specific contaminants in sediment and biota facilitated a discussion on potential exposure, independent of particle accumulation in the gut. One of the primary objectives of the investigation was to determine the relative importance of local and remote sources for plastic contamination in the Arctic, and preliminary results indicate a clear signal from local sources and sea ice. In order to evaluate the risk posed by microplastics in the Arctic, a risk analysis already burdened by multiple stressors, knowledge about sources, fate and concentration of microplastics in different environmental compartments is crucial.

TH032
Microplastics – an ecotoxicological issue? How to balance facts and perception without marginalizing an environmental problem

C. Völker, ISOE - Institute for Social-Ecological Research / Water infrastructure and risk analyses; J. Kramm, ISOE - Institute for Social-Ecological Research

Microplastics have been known as a factor for environmental pollution – symbolized by the plastic bag – for a long time, recent scientific evidence on the massive accumulation in the oceans and the environmental risks associated with microplastics in the Arctic, a risk already burdened by multiple stressors, knowledge about sources, fate and concentration of microplastics in different environmental compartments is crucial.

TH033
Assessing biotransformation and bioconcentration factors (BCF) of fragrance materials using in vitro approaches utilizing rainbow trout liver S9 sub-cellular fractions and cryopreserved hepatocytes

A. Łapczynski, RFIM / Environmental Science; K.M. Johanning, KJ Scientific LLC / dba of Pura Vida Connections LLC; A. Jenkins, EAG Laboratories

Bioaccumulation potential measured as the bioconcentration factor (BCF) is one of the components for the PBT (persistent, bioaccumulative, toxic) criteria and risk assessment of chemicals by regulatory agencies in some regions of the world (e.g. REACH and ECHA). Biotransformation and metabolism of chemicals in the environment are a source of uncertainty in bioaccumulation assessment. Currently, in vitro methodologies utilizing the rainbow trout metabolic assay not only are gaining interest, but are being used increasingly by several sectors as a crucial component in model-based estimates of BCFs and as part of a line of weight of evidence presented to regulators. The rainbow trout screening assay utilizing liver S9 fractions and cryopreserved hepatocytes to test chemical biotransformation has gone through a Ring Trial for an OECD validation process (OECD Project 3.13 coordinated by ILSI HESI) and the Test Guidelines and Guidance are being reviewed by the OECD assigned panel. In the present study four fragrance materials (Cyclabute, Melafleur, Trimofix and Verdox) with known measured BCF values obtained using OECD 305 method were tested for biotransformation utilizing both rainbow trout liver S9 fractions and cryopreserved hepatocytes. The results indicate that all four fragrance materials were metabolized in both biological systems at different rates, but in all cases the BCFs determined were comparable to the measured in vivo BCF values. The in vitro metabolic assay is a powerful tool that can be used to determine BCF of test chemicals and provide data to build the database information on fragrance materials for fish metabolism and modeling.

TH034
Addressing species diversity in biotransformation: variability in expressed transripts of hepatic biotransformation enzymes among fishes


There is increasing evidence that diverse xenobiotic metabolizing enzymes exist among fishes, potentially resulting in different chemical sensitivities and accumulation, but this has never been systematically evaluated. One concern is that model test species such as rainbow trout, zebrafish and fathead minnows may not adequately represent the xenobiotic metabolizing capacity of other fish species. Our current study mined available fish liver transcriptome data for expressed enzymes utilising full-transcript, isoform sequencing on liver samples from two dozen phylogenetically diverse fish species. This novel RNAseq approach eliminated the need for transcriptome reconstruction resulting in reference genomes of the highest precision, allowing for detection of enzyme isoform orthologs among the species, as well as the nuclear receptors that control expression of the enzymes. Species including dealkylation, hydroxylation, carboxylation, oxidative dehalogenation and brominated flame retardants (BFRs), are commonly detected in environmental samples and assessed for biotransformation utilizing both rainbow trout liver S9 fractions and cryopreserved hepatocytes. The resolved isoforms were compared to the measured in vivo BCF values. The content of this presentation neither constitute nor necessarily reflect US EPA assessments or metabolism prediction software for potential relevance in fish.

TH035
Metabolism of Organophosphate Flame Retardants (OPFRs) in Freshwater Fish: Field and Laboratory Studies

R. Hou, X. Li, R. Hou, Research Center for EcoEnvironmental Sciences, Chinese Academy of Sciences / Key Laboratory of Drinking Water Science and Technology; D. Feifarek, SCJohnson; D.L. Villeneuve, U.S. EPA / National Health and Environmental Effects Research Laboratory

There is increasing evidence that diverse xenobiotic metabolizing enzymes exist among fishes, potentially resulting in different chemical sensitivities and accumulation, but this has never been systematically evaluated. One concern is that model test species such as rainbow trout, zebrafish and fathead minnows may not adequately represent the xenobiotic metabolizing capacity of other fish species. Our current study mined available fish liver transcriptome data for expressed enzymes utilising full-transcript, isoform sequencing on liver samples from two dozen phylogenetically diverse fish species. This novel RNAseq approach eliminated the need for transcriptome reconstruction resulting in reference genomes of the highest precision, allowing for detection of enzyme isoform orthologs among the species, as well as the nuclear receptors that control expression of the enzymes. Species including dealkylation, hydroxylation, carboxylation, oxidative dehalogenation and brominated flame retardants (BFRs), are commonly detected in environmental samples and assessed for biotransformation utilizing both rainbow trout liver S9 fractions and cryopreserved hepatocytes. The resolved isoforms were compared to the measured in vivo BCF values. The content of this presentation neither constitute nor necessarily reflect US EPA policy.
the accumulation and tissue distribution of eight common OP FRs and their four DAP metabolites in three freshwater fish species from locations around Beijing, China. Accumulation of DAPs were relatively lower but comparable to those of PFRs in freshwater fish. DAPs had low limited affinity to lipid content in tissues, similarly like their parent compounds PFRs. Liver was identified to have a higher accumulation of PFRs and DAPs than the other tissues of fish. It suggested the expressed xenobiotic-induced P450s in wild animal studies. In the consequent laboratory control study, we screened the metabolites of alkylo-PFRs by in vivo exposure of Gobiocephalus rarus. Metabolites of alkylo-PFRs in fish liver after 30-day exposure were analyzed with UPLC-QTOF/MS in MSE mode. The qualitative results verified the metabolic pathway of dealkylation, hydroxylation, dihydroxylation, desaturation, and phase II glucuronide conjugation for all the tested three alkylo-PFRs. We identified and accurately quantified the metabolites 3-OH-TBOEP, BBOEHEP, and 3-OH-TNBP formed in fish liver microsomes. Liver rather than intestine, plays the primary role in PFR clearance in fish. The significance of these metabolites is in good agreement with human urine monitoring and in vivo rat exposure studies. Overall, the results emphasized the importance of hydroxylated metabolites as biomarkers for alkylo-PFRs exposure.

TH036 Bioaccumulation and biotransformation of prochloraz in the aquatic invertebrate Hyalella azteca

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Prochloraz is a widely applied fungicide for pest management purposes. Due to spray drift and surface runoff, prochloraz enters the aquatic environment where it can be a risk to individual species. Prochloraz has been recognized as a model organism to test toxicity of organic chemicals due to its rapid life cycle, the feasibility of cultivation and its sensitivity to xenobiotics. Biotransformation is a primary detoxification process through which organisms defend themselves from xenobiotics. Biotransformation can reduce the internal concentration of parent compounds hence, influencing their bioaccumulation. The aim of this study was to assess the toxicoxicokinetics of prochloraz and its biotransformation products (BTPs) in Hyalella azteca. Adults of Hyalella azteca were exposed to prochloraz at the concentration of 100 µg L⁻¹ during a 24-hour uptake phase and a subsequent 120-hour depuration phase. Organisms were sampled over time and after extraction, the internal concentration of prochloraz and its BTPs were quantified using reverse phase liquid chromatography coupled to high resolution mass spectrometry with electrospray ionization. Prochloraz and its 30 BTPs were detected, quantified, and respective toxicokinetic profiles were obtained. In every profile, an increase in the internal concentration was seen during the uptake phase followed by a decrease during the depuration phase. The bioaccumulation factor was calculated to be 110 Lkg⁻¹. Finally, the data will be modeled using toxicokinetic analysis. An elimination and transformation rate constants will enable determining the role of biotransformation in the detoxification of prochloraz in Hyalella azteca.

TH037 Toxicoxicokinetics and metabolite identification of two emerging pollutants, Alkyl-substituted 4- and 3MBC, in the manila clam Ruditapes philippinarum.

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Marine ecosystems have been historically sinks for many pollutants and chemicals whose effects awoke social concern, triggering the implementation of legislations. Nowadays, new compounds are developed at increasing rates and eventually discharged into marine ecosystems in unknown quantities and with no regulation. Due to the improvement of new analytical techniques, many of these chemicals, the so-called "emerging pollutants" (EPs), are being currently identified and their occurrence is being proven in the environment. However, very little is known about the possible adverse effects of these emerging pollutants in exposed non-target organisms. In this context, the present work evaluates the toxicoxicokinetics (TK) of two EPs (the UV filter 4-Methylbenzylidene-camphor (4-MBC) and the artificial sweetener ascesulfame K (ACE-K)) in the Manila clam Ruditapes philippinarum, focusing on determining the bioaccumulation factors (BCF) and identifying and quantifying the metabolites and products found. After 7 days of exposure and 3 days of depuration, target compounds were extracted from both water phase and organisms and their concentrations were measured by liquid and gas chromatography coupled to tandem mass spectrometry (UPLC/QC-MS/MS). Additionally high resolution mass spectrometry (HRMS) and automated data analysis software (Metabolynx™) were used to identify possible TPs in the tissue of the Manila clam at different nominal concentrations (from 1 to 100 µg L⁻¹). For the UV filter, the estimated BCFs were between 61 553 and 539 133 L kg⁻¹, and several metabolites were identified, such as the reduction or hydroxylation of the compound. On the other hand, the artificial sweetener BCF was consistently lower, around 7 L Kg⁻¹ and no metabolites were identified. These results suggest that 4-MBC was highly bioaccumulated and metabolized to facilitate its excretion and they are directly related to the physicochemical properties of the target EPs, since ACE-K is highly soluble in water (log Kow, 1.33) and excreted unchanged in comparison to the very hydrophobic UV filter (log Kow, 5.92). Additionally, the present study provides important information about the toxicity of EPs, 4-MBC and ACE-K, which will be helpful for understanding the mechanism of action of these compounds. Furthermore, this work demonstrates the potential of the UPLC-GC/HRMS approach using Metabolynx™ software for fast and accurate identification of metabolites of EPs.

TH038 Organophosphate Esters, Including Alkyl-Substituted Triphenyl Phosphates, in East Greenland Polar Bears and Ringed Seals: A rejoice Tissue Concentrations and In Vitro Depletion and Metabolite Formation

A. Strobel, Carleton University; W.G. Willmore, Carleton University / Biology department; C. Sonne, R. Dietz, Aarhus University / Department of Biosciences, Arctic Research Centre; R. L. Letcher, Environment and Climate Change Canada / Ecotoxicology and Wildlife Health Division

After the global decrease in the use of organophosphate esters (OPEs), they were identified as contaminants of particular concern in marine ecosystems. Marine animals, including seals and polar bears, have been identified as biota that accumulate these contaminants in their tissue. The study aimed to assess the toxicokinetics of prochloraz and its biotransformation products (BTPs) in Hyalella azteca. Adults of Hyalella azteca were exposed to prochloraz at the concentration of 100 µg L⁻¹ during a 24-hour uptake phase and a subsequent 120-hour depuration phase. Organisms were sampled over time and after extraction, the internal concentration of prochloraz and its BTPs were quantified using reverse phase liquid chromatography coupled to high resolution mass spectrometry with electrospray ionization. Prochloraz and its 30 BTPs were detected, quantified, and respective toxicokinetic profiles were obtained. In every profile, an increase in the internal concentration was seen during the uptake phase followed by a decrease during the depuration phase. The bioaccumulation factor was calculated to be 110 Lkg⁻¹. Finally, the data will be modeled using toxicokinetic analysis. An elimination and transformation rate constants will enable determining the role of biotransformation in the detoxification of prochloraz in Hyalella azteca.

TH039 Proteomics of a metabolic simulation system - a look inside rat S9

A. Schiwy, EWMOS; B. Thalmann, RWTH Aachen University, Institute for Environmental Research / Bio5 - ESA; P. Huesen, Forschungszentrum Jülich GmbH / Central Institute for Engineering, Electronics and Analytics (ZEA); Schiwy, Institute of Environmental Research-RWTH Aachen / Department of Ecosystem Analysis; H. Bollert, RWTH Aachen University / Institute for Environmental Research

The liver is the key organ in metabolism and detoxification of xenobiotics. Simulation of this organ in various bioassays is achieved via the application of either single cytochrome P450 enzymes produced via biotechnological processes or complex enzyme mixtures obtained from animals. Especially, this second process is regarded under various animal welfare regulations as an animal experiment. The animals have to be maintained to a specific age and following their lives have to be induced via various chemicals. Furthermore, this treatment may cause pain to the animals. Finally, the animals have to be killed to harvest the organ (predominantly liver) for further downstream processing. The most common procedure is a mincing of the organ and separation of subcellular fractions. In vitro OP triester metabolism was generally rapid and structure-dependent, where PBs metabolized OPEs more rapidly than RSs. Exceptions were the lack of triethyl phosphate metabolism and slow metabolism of triis (2-ethylhexyl) phosphate in both species. OP diester metabolites were also formed with the exception of triphenyl phosphate (TPHP) which was not metabolized at all in the RS assessment. Tri-(3-dichloro-2-propyl) phosphate was completely converted to its corresponding diester. The mass balances showed that OP diester formation corresponding to triis (2-ethylhexyl) phosphate, tri (n-butyl) phosphate, and tri (2-butoxyethyl) phosphate did not account for 100 % of the OP triester depletion, which indicated alternate pathways of OP triester metabolism. TPHP was completely converted to its OP diester metabolite in PBs but not in RSs suggesting species-specific differences. Alkyl-substituted TPHP analogues also showed that the number and position of the phenyl ring substitution heavily influenced the rate of metabolism. The results demonstrated that OP triester bioaccumulation and fate in PBs versus their RS prey is substantially influenced by biotransformation.
mammals and fish
J.A. Arnot, ARC Arnot Research & Consulting / Adjunct Professor, Department of Physical & Environmental Science/Department of Pharmacology and Toxicology; A. Looky, ARC Arnot Research and Consulting Inc.; K.L. Foster, ARC Arnot Research and Consulting Inc. / Adjunct Professor, Trent University, Applications of Modelling & Quantitative Methods (AMOD); J.M. Armitage, University of Toronto - Scarborough / Physical and Environmental Sciences; M. Halder, European Commission Joint Research Centre / DG Joint Research Centre IHC EUR LCVAM; A. Lostia, European Commission Joint Research Centre; A. KIENZLER, JRC-EC / F3-Chemical Safety and Alternative Methods Unit IHC EUR LCVAM

Toxicokinetics (TK) plays an important role in ecological and human health assessments. To this end, we compared TK data on chemicals to subject regulatory assessment requirements. It is not feasible to measure TK data in all organisms (e.g., humans, rodents, fish) and there is a recognized need to reduce animal testing. Reliable (evaluated), high-quality existing in vitro and in vivo TK data could help evaluate in vitro in vivo extrapolation models (IVIVE), parameterize TK and bioaccumulation models, and develop and validate quantitative structure-activity relationships (QSARs) for predicting TK parameters from chemical structure. Biotransformation and elimination rate data can be used in diverse contexts for chemical assessment. For example, biotransformation rate constants (k_B) are key determinants and sources of uncertainty in bioaccumulation assessment. k_B can be determined in vivo with whole animal models or from in vivo assays using intact cells or subcellular fractions from the liver or other tissues (e.g., gastrointestinal tract, kidney). We have developed a new database (funded by the JRC CCR.F.C93136.X0) containing TK data (i.e., biotransformation rates) for fish and mammal species (i.e., rat, mice) derived from in vivo and in vitro (S9 fraction, hepatocytes, microsomes) methods. The database entries are scored based on a data quality evaluation. The data quality assessment methods and criteria have been developed from standardized testing guidelines when such guidance exists and from professional judgement in the absence of standardized guidance. In total the new database includes approximately 9000 entries for organic chemicals. There are approximately 4000 and 400 chemicals from in vitro and in vivo studies respectively from rodent species. There are approximately 120 and 700 chemicals from in vitro and in vivo studies respectively from fish species. The database can be used as a source of information for chemical assessments and can help identify future research needs (i.e., chemicals that require chemical evaluation and for which reliable quality data are not available). We believe the database will also be a valuable source information for model developers (e.g. for in vitro-in vivo extrapolation models, kinetic models, models to predict exposure and internal concentration in an organism) and chemical evaluators. The database will be publicly available at the Joint Research Centre website.

TH041
A tiered testing strategy for rapid estimation of bioaccumulation by a combined modelling - in vitro testing approach: derivation of kinetic rate constants
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Our research seeks to improve alternative methods to estimate bioaccumulation of organic chemicals in fish. We follow a tiered strategy that integrates toxicokinetic (TK) models, quantitative structure-activity relationships (QSARs), in vitro exposure routes, and tissue collection (i.e., gill and trout), and in vivo in vivo extrapolation methods. In a first step, we derived a set of candidate chemicals for in vitro testing based on model discrepancies, availability of reliable in vivo BCF and BMF data, and availability of in vitro biotransformation rates. The resulting chemicals were divided into three K_B categories based on predominant exposure route(s) to guide in vitro testing: 1) log K_B > 4 (aqueous exposure dominates – to be tested in gill and liver models); 2) 2.5 < log K_B < 5.5 (mixed exposure routes – to be tested in gill, liver and intestine models); and 3) log K_B < 2.5 (predominantly dietary exposure dominates – to be tested in liver and intestinal models). In vitro testing is now on-going. Specifically, primary gill cell cultures grown on permeable support are used to determine the combined biotransformation/permeation rate through this epithelial model. Primary suspension preparations from hepatocytes and intestine are explored for biotransformation rates (i.e. loss of parent compound). Permanent cell lines of gills, liver and intestine, exposed in monolayer, complement the set of in vitro methods applied, yielding parent compound loss rates as well. In vivo models are each applied under their respective optimal conditions, taking e.g., temperature and media composition into account. Chemical starting concentrations are set uniformly for all models based on non-toxic concentrations and analytical method sensitivity. Thus far, permeation/biotransformation was observed for all chemicals applied. The resulting rate constants are subject to comparison between the different in vitro model sets and are input into the TK and QSAR models for model development and hypothesis testing. This poster will describe the overall in vitro testing strategy, the different in vitro models and the results of the chemical testing with regard to in vitro-derived rate constants.

TH042
Update on development of OECD Test Guidelines and Guidance Document on determination of fish in vitro hepatic clearance

Chemical biotransformation represents the largest source of uncertainty in chemical bioaccumulation assessments, and model-based estimates of chemical bioconcentration in fish may be greatly improved by including biotransformation rates, as measured in vitro. Substrate depletion assays, using rainbow trout hepatocytes (RT-HEP) or liver subcellular fractions (RT-S9), have been successfully developed and optimized to model rates of fish biotransformation. A two-tier, multi-laboratory ring trial, coordinated by the ILSI Health and Environmental Sciences Institute (HESI), was recently completed which demonstrates assay reliability within and across laboratories and similar performance of substrate depletion assays using the two biological systems. Based on the successful results of this ring-trial, two OECD test guidelines (TG) (“Determination of in vitro intrinsic clearance using cryopreserved rainbow trout hepatocytes” and “Determination of in vitro intrinsic clearance using rainbow trout liver S9 sub-cellular fractions”) have been drafted and are accompanied by a Guidance Document (GD). The OECD GD provides detailed information on how to conduct the tests as well as how to apply the measured in vitro biotransformation rates to predict bioconcentration factors. In addition, guidance on selection of the assay system (e.g., primary hepatocytes vs. liver S9 fractions), specific considerations for testing chemicals, use of negative and positive controls, BCF extrapolation models, and application of the two test methods beyond BCF prediction are also covered. Draft TGs, the GD, and the ring trial report underwent two OECD public commenting rounds during 2017 and submission to OECD WNT final approval is planned for 2018.

TH043
The Bioaccumulation Assessment Tool (BAT): A quantitative weight of evidence approach for bioaccumulation assessment
L. Toose, ARC Arnot Research & Consulting; J.M. Armitage, University of Toronto - Scarborough / Environmental Toxicology; J.A. Arnot, ARC Arnot Research and Consulting Inc. / Adjunct Professor, Trent University, Applications of Modelling & Quantitative Methods (AMOD); M. Embry, ILSI; J.A. Arnot, ARC Arnot Research & Consulting / Adjunct Professor, Department of Physical & Environmental Science/Department of Pharmacology and Toxicology

Chemicals are being assessed for bioaccumulation (B) potential in regulatory programs using various methods, metrics and criteria. B data can be obtained from various data streams including laboratory studies, field studies and model predictions using mass balance models and quantitative structure-activity relationships (QSARs). Examples of bioaccumulation metrics include: the bioconcentration factor (BCF), bioaccumulation factor (BAF), biomagnification factor (BMF), trophic magnification factor (TMF). The Bioaccumulation Assessment Tool (BAT) to collect, evaluate and integrate various lines of evidence (LOE) associated with these B-metrics and related B classification criteria to aid decision-making. The BAT provides a transparent and consistent framework for evaluating neutral and ionizable organic chemicals in aquatic and terrestrial organisms. It uses a quantitative a weight of evidence (QWOE) approach which includes evaluations for the relevance, reliability (confidence) and outcome of each B-metric. Each substantive LOE (e.g., BCF, BMF, biotransformation rate) is subject to data quality evaluation resulting in a data confidence score. The Data Evaluation Templates (DETs) have been derived from standard test protocols and expert judgment when standard protocols are not yet developed. Physical-chemical properties can be used or the user is allowed to enter biologically relevant partition coefficients in place of default assumptions that assume octanol as surrogate for biological components (i.e., lipid). Estimates for biotransformation rates can be included from in vitro assays (i.e., S9, hepatocyte, microsomal) and from in silico (QSAR) predictions. Empirical data such as lab BCFs and BMFs and field data as

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well as in silico data (e.g., BCF-QSARs) can be included in the QWOE. This presentation provides an overview of the BAT and demonstrates its application with two case studies. The first example is a typical “data poor” scenario in which only chemical structure information is available. From chemical structure relevant physical-chemical property and biotransformation rate data are obtained from QSARs and entered into the system. The second case study is for a relatively data rich set of the EU EED (e.g., 3 lab BCFS, various BCF-QSARs, biotransformation rate QSARs, in vitro biotransformation rates). Future work for improving the BAT is discussed.

TH044 Towards the use of elimination rates in bioaccumulation assessment – Current challenges and future trends

G. Treu, German Environment Agency / REACH Chemicals; W. Drost, Federal Environment Agency (UBA) / Chemicals; C. Rauert, Umweltbundesamt / International Chemicals Management

The capacity of chemicals to bioaccumulate in biota is recognized as critical property that contributes to a chemicals risk. The bioconcentration factor (BCF) reflecting the uptake of a chemical from water and the biomagnification factor (BMF) following dietary uptake in fish remain the preferred metrics in bioaccumulation assessment. The test systems are expensive, time consuming and are not suitable for screening purposes. Still, terrestrial bioaccumulation is hardly considered. A comprehensive bioaccumulation assessment should consider both, the aquatic and terrestrial organisms. Recently, it has been suggested that BCF and BMF can be derived by only determining the elimination rate constant ($k_1$) experimentally while the uptake rate ($k_2$) is estimated. Following this concept the need for animal tests is reduced if the metabolic contribution to $k_2$ is from $in vitro$ experiments while the effect of the other pathways (excretion via urine and feces, and ventilation) are estimated with $in vitro$ to vivo extrapolation models. Biotransformation often reduces the extent to which chemicals accumulate in fish and mammals. Thus, a Tier 1.5 can be introduced between Tier 1 ($screening based on physico-chemical data$) and Tier 2 ($exposure studies with animals where $in vitro$ biotransformation rates ($k_3$) obtained from $in vitro$ tests with fish or mammalian cells are extrapolated to whole organisms and then incorporated into to existing chemical mass-balance models to predict a BCF or BMF. Only if this model indicates an increased bioaccumulation a potential a higher-tier vertebrate test is then needed. In practice, animal tests are mandatory for chemicals exceeding a certain level of hydrophobicity but may turn out as non bioaccumulative due to metabolisation. A $k_2$ based extrapolation model allowing to estimate BCF and BMF values by incorporating $in vitro$ $k_3$ of different tissues, e.g. gills, liver and gastrointestinal tract, could be serve as alternative screening criterion under REACH. This model could further cover species differences currently ignored in bioaccumulation regulation. However, uncertainties remain related to the validity of this approach, e.g., for iontotoxic substances, and should be addressed in future research by taking into account specific metabolic pathways. This poster aims at demonstrating current limitations and future needs for the $k_3$ based bioaccumulation assessment under REACH from a regulatory agency’s perspective.

TH045 SETAC Bioaccumulation Science Interest Group

L.P. Burkhard, U.S. EPA / ORD/NHEERL/Mid-Continent Ecology Division

Advances in evaluating and regulating endocrine disruptors (P)


K. Yamazaki, Ministry of the Environment / Environmental Health Ministry of the Environment, Japan published its fourth program on endocrine disrupting effects of chemical substances “EXTENDED2016” in June 2016. It is developed upon achievements on development of framework, development and improvement of test protocols and implementation of testing and assessment in the preceding program “EXTENDED2010”. While basic concepts and framework was inherited from EXTENDED2010, EXTENDED2016’s focus has been shifting to implementation of testing and assessment and consideration of appropriate risk management measures. During fiscal years 2016-17 progress has been made in development of test protocols, evaluation of existing knowledge, identification of candidate chemicals for testing, implementation of testing and assessment and communication to the public, as well as in international collaborative projects with the United Kingdom and the United States. One of the most significant achievements should be finalization of the data obtained from the medaka extended one generation reproduction test (MEOGRT) for 4-nonylphenol, which are expected to be referred to in regulatory environmental risk assessment. The reproduction tests are being conducted for additional chemicals within the program. Updated progress in testing and assessment under EXTENDED2016 will be presented at the Annual Meeting.

TH047 Effects of endocrine disruptors on reproductive health: A new approach to integrating ecotoxicological and human health data

L. Parent, Télé-université / UER Science et Technologie; P. Grigorova, Université TELUQ / Département Science et Technologie; M. Nikolaraos, Université TELUQ / Science et Technologie

Exposure to synthetic and natural chemicals is almost inevitable in our daily lives. Some of them raise concerns with their endocrine disruptive potential and possible interference with endocrine system leading to the variety of adverse health effects. It was initially through clusters of presumptions that the potential effects of endocrine disruptors (EDs) on human health and the environment were highlighted. EDs, as a growing source of concern, now need to better document the complexity of a relationship between exposure and effects, hence the development of new evidence-based approaches to better document decision-making in health policy. Among these approaches, we retained the systematic reviews, based on objective methods, to integrate multiple sources of evidence (epidemiology, wild animals, laboratory animals, in vitro and in silico data) relevant to the evaluation. Our project aims to systematically review the data published the last 10 years linking the existing to EDs (polybrominated diphenyl ethers (PBDE), alkylphenols, bisphenol A (BPA), parabens, phthalates, perfluorinated compounds) with the effects on the development and reproductive health as changes in sex ratio, congenital malformations, sperm quality disruption, alteration of plasma levels vitellogenin, sex hormone levels as well as anomalies of gonad development. 16 701 articles were screened and 744 met the inclusion criteria for the review. The data was extracted from 155 EU legal frameworks, 542 fish tests, 377 ecotoxicological studies and the ROB (risk of bias) analysis was performed for the relevant outcomes, confidence in the body of evidence for an effect was rated, and scores are given. In this presentation, we will show what is the strength of the evidence for the association between exposures and (adverse) effect, and we will discuss the role of ecotoxicological studies in the global analysis: prioritizing EDs, understanding mechanisms of action, establishing standards or impact criteria, identifying sensitive biomarkers and bioindicators for each of the EDs.

TH048 Pros and cons of fish toxicity tests in detecting chemicals with endocrine disrupting activities

A. Kienzler, JRC-EC / F3-Chemical Safety and Alternative Methods

Unit-EURL ECVM; Z. Dang, RIVM / LIEC CNRS UMR; S. van der Linden, JRC-EC

In the last decade, with the increased awareness of potential effect of endocrine disruptors (EDs) on wildlife, fish toxicity test guidelines have been developed by administrations and expert committees. The aim of these guidelines is to identify EDs to identify chemicals with estrogenic and anti-estrogenic activities and to validate these guidelines. These activities have been summarized in the OECD guideline documents 150 and 171 and specific guidance on the diagnosis of endocrine-related histopathology in fish gonads is available (GD 123). However, while the relevance of fish toxicity tests is clear in the assessment of endocrine disruptors, comparison of these tests in response to EDs has not yet been made in three different areas: in vitro, in vivo and in fish tests. In this study, the relevance of these fish tests are evaluated on the basis of the existing information case by case. Due to a difference in sensitivity of species and life stages, many EU discussions have focused on which test should be suitable. However, due to practical considerations, (e.g. regional preference, practical use, and specifically sensitivity,) it is unlikely to meet all of the requirements within one test. But in order to avoid further additional testing, species sensitivity should always consider the best factors. This work intends to summarise the pros and cons of the available test guidelines and to address some issues e.g. sensitivity in different life stages and in species. Available fish toxicity tests include test guidelines (TG) 229, 230, 234, 240 and guidance document (GD) 148. The number of fish used in each fish test, the covered lifestage, the investigated EDs-related endpoints, their robustness (and to which extend these have been validated) and the species sensitivity in response to chemicals with EAS modes of action will be compared. To this aim, publically available data on different fish species tested according to TGs or TG-like protocols will be collected and analyzed. Analysing these fish toxicity data will help identifying which fish test, which species, which life stage of test are needed for the identification and/or risk assessment of EDs. Based on the overall data analysis, we will propose an environmental testing strategy, which is important for minimizing vertebrate testing and costs.

TH049 Towards developing a list of reference chemicals for endocrine assay validation

C. Prosser, ExxonMobil Biomedical Sciences, Inc.; M.R. Embry, ILSI Health and Environmental Sciences Institute (HESI)

Compared to other areas of human and environmental hazard assessment, evaluating the potential for endogenous compounds to interact with endocrine pathways is relatively nascent. However, recognizing the possibility of a growing workforce of EDs, many national governments, international organizations, industry bodies, public interest groups and academic institutions established research programs to address the impacts of endogenous substances on the endocrine system. This has resulted in attempts to develop and validate a battery of
tests to screen for endocrine active compounds with multiple publications by both regulatory agencies and academics aimed at identifying appropriate \textit{in vitro} and \textit{in vivo} assays. Thus, there has been considerable effort to establish criteria and interpret results for the identification of potential of endocrine active compounds. However, despite all the attention on test development, little consideration has been given to establishing a list of reference compounds to be used in the validation process. Without establishing a set of criteria it may prove problematic to assess interlaboratory variability for the same endocrine mode of action (e.g. estrogenic/androgenic effects). When evaluating the current, validated, assays in OECD Guidance Document 150, there is a great disparity in the reference chemicals selected and, no discussion as to why various chemical were chosen for the validation procedure. Additionally, reference chemical selection is often not communicated with the corresponding ring trials. While these TGs focus on a diverse range of toxic effects, none of their (unintentional) toxicity and approve their use. In case of concern(s), specific research is needed.

Some well accepted and commonly used chemicals are provided as a realistic starting point to compile a list of reference chemicals for the validation of endocrine assays.

**TH050**
Assessment of endocrine disrupting properties of pesticides and biocides: data processing to support data analysis
A. Lostia, Joint Research Centre - European Commission - Institute for Health and Consumer Protection; S. Munn, European Commission; S. van der Linden, European Commission Joint Research Centre / Directorate F Health, Consumers and Referen Material; A. Kienzler, JRC-EC / F3-Chemical Safety and Alternative Methods Unit-EURL ECVAM; E. Grignard, European Commission Joint Research Centre; E. Joosens, European Commission DG Joint Research Centre

In 2016 the Commission completed an impact assessment to estimate possible health, environmental and socio-economic consequences of adopting different options, formulated as scientific criteria, for identifying Endocrine Disruptors (EDs) under the Plant Protection Products Regulation and Biocidal Products Regulation. JRC developed a central element of this assessment, namely, a science-based methodology to screen over 600 chemicals in about 10 months, including all EU-registered biocides and pesticides. The methodology was based solely on already existing data. To achieve the objective of screening hundreds of substances in a limited time, all retrieved data for a substance (toxicological studies, effects observed, NOAEL, etc.) are captured in an excel template (consisting of 40 columns), developed by JRC, in order to systematically organise the information to then facilitate data-analysis. As a result a large and curated database is available summarising relevant existing data collected for the 600 substances screened. A major accomplishment was the development of an innovative way to process and visually represent the data captured in the excel template as a mean to facilitate the data analysis in a systematic manner and in medium-throughput to ensure meeting the objective defined in the Commission Roadmap of screening 600 substances in a limited time and by using high-quality science-based strategy. Briefly the data collected are re-organised and processed into a data-matrix which is built automatically after the template has been filled. The data-matrix, available for each of its components, is a powerful tool for getting access to toxicological information available for a certain chemical thus facilitating the data analysis to identify EDs. For instance the data-matrix visually reports if certain toxicological endpoint has been observed across different studies to support evaluation of the biological plausibility of a causality or mechanism of action (MoA). Under certain legislations, understanding the potential MoA is particularly important because regulatory decisions might categorise a substance as an endocrine disruptor (ED) through a hazard characterization process rather than taking into consideration exposure and risk. The WHO/IPCS definition of an endocrine disruptor requires that a substance alter the functions of the endocrine system of the intact organism. Pathway models are being established that provide plausible links between molecular initiating events, key events and ultimately adverse effects. However, when several potential pathways or MoAs converge on the same adverse effect in an intact organism it becomes challenging to identify the biologically plausible causal link between the MoA and the environmental or health effect of regulatory concern. The first part of the WHO/IPCS definition, that of the chemical acting through an endocrine MoA to alter the function of the endocrine system, focuses on the need for the substance to have sufficient affinity for and activity with components of the endocrine system, compared to endogenous hormones, in order to compete with the normal hormonal signaling and feedback mechanisms that maintain homeostasis. Additionally, some well accepted and commonly used chemicals are provided as a realistic starting point to compile a list of reference chemicals for the validation of endocrine assays.

**TH052**
Plausible or Causal: Bioactivity and mechanistic potency as a critical piece in hazard characterization of endocrine active chemicals
E. M. Stühler, E2; K. Plötzer, Dorn Chemical Company / Toxicology, Environmental Research & Consulting
While methods have been and are being developed and validated, and regulatory programs around the world are moving forward with evaluating chemicals for their potential interaction with the endocrine system of humans and wildlife, the challenge still remains in distinguishing between effects that are specifically exogenously mediated from those that are a result of general stress or some other mode or mechanism of action. Under certain legislations, understanding the potential MoA is particularly important because regulatory decisions might categorize a substance as an endocrine disruptor (ED) through a hazard characterization process rather than taking into consideration exposure and risk. The WHO/IPCS definition of an endocrine disruptor requires that a substance alter the functions of the endocrine system of the intact organism. Pathway models are being established that provide plausible links between molecular initiating events, key events and ultimately adverse effects. However, when several potential pathways or MoAs converge on the same adverse effect in an intact organism it becomes challenging to identify the biologically plausible causal link between the MoA and the environmental or health effect of regulatory concern. The first part of the WHO/IPCS definition, that of the chemical acting through an endocrine MoA to alter the function of the endocrine system, focuses on the need for the substance to have sufficient affinity for and activity with components of the endocrine system, compared to endogenous hormones, in order to compete with the normal hormonal signaling and feedback mechanisms that maintain homeostasis. Additionally, some well accepted and commonly used chemicals are provided as a realistic starting point to compile a list of reference chemicals for the validation of endocrine assays.

**TH053**
Addressing endocrine concerns for the environment in dossier evaluations with an FDST – possibility to avoid further vertebrate tests
F. Kaßner, German Environment Agency UBA; S. Germer, German Environment Agency UBA; E. Hassold, German Environment Agency UBA / IV 2 Chemicals; S. Munn, European Commission DG Joint Research Centre; E. Grignard, European Commission Joint Research Centre; A. Kienzler, JRC-EC / F3-Chemical Safety and Alternative Methods Unit-EURL ECVAM; E. Joosens, European Commission DG Joint Research Centre

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TH054 Structural Alerts for Potential Endocrine Disruptors
R. Kühn, N. Ost, Helmholtz Centre for Environmental Research UFZ / Department of Ecological Chemistry; L.A. Baumann, University of Heidelberg / Animal Ecology and Toxicology; H. Seggewiss, W. Arlt, Helmholtz Centre for Environmental Research UFZ / Centre for Fish and Wildlife Health; J. Arning, German Environment Agency UBA / Chemicals; G. Schuurman, Helmholtz centre for environmental research - UFZ / Department of Ecological Chemistry

Endocrine disrupting chemicals interact with the hormone system. They may trigger adverse effects on organisms. Endocrine disrupters are labelled as substances of very high concern (SVHC) and are subject of regulations as REACH. However, there are so far no internationally harmonised criteria for endocrine activity. Furthermore, the endocrine system is rather diverse. Existing tests are rather expensive, and it is still not clear whether they comprise all relevant pathways. Thus, the number of existing data is limited. In silico tools may provide alternatives at least to allow prioritisation of tests by screening compound lists. This study aimed at identifying structural alerts for potential endocrine disruptors of two relevant hormone systems, estrogen/androgen and thyroid hormones. Chemicals binding to the estrogen/androgen receptors may either yield an agonistic effect by mimicking the hormone, or an antagonistic effect by blocking the receptor site and thus preventing the hormones from binding themselves. Thyroid hormones bind to the ligand binding domain (LBD) of the receptor, and secondly binding of a co-activating protein to a part of the LBD (AF-2) triggers gene expression. Chemicals binding to LBD may again yield agonistic or antagonistic effects, for binding to AF-2 only antagonism is known. However, adverse effects to thyroid hormones can also result from other mechanisms as enzyme/protein interaction e.g. with the transporter protein and aryl-hydrocarbon-receptor interaction. Structural alerts to predict chemicals with potential effect on these systems have been developed. For the estrogen/androgen systems the approach has been refined. The model identifies 91% of the active chemicals, and false negative results are weakly active only. The models for LBD and AF-2 binding only miss one active compound. For other thyroid hormone effects a screening level model detects ca. 95% of the known active compounds, but there is suspect of missing compound classes due to the lack of respective experimental data. Particular remark was given to characterize the applicability domain and reliability of the predictions. All models are implemented as automated tools in the software system ChemProp (UFZ Department of Ecological Chemistry 2017. ChemProp 6.6. http://www.ufz.de/ecochem/chemprop). Acknowledgment: This study was financially supported by the German Federal Environment Agency, FKZ 371463412 (O).

TH055 Mixtures of endocrine disrupting chemicals disrupt behaviour and thyroid hormone related gene expression in Zebrafish (Danio rerio) larvae
L. Bjergersson, J. Sturve, University of Gothenburg / Department of Biological and Environmental Sciences

Endocrine disrupting chemicals (EDCs) in the aquatic environment can have severe effects on the health of aquatic organisms as well as human health. Numerous anthropogenic EDCs, such as plasticizers, fire retardants and antibacterial agents, enter aquatic ecosystems from wastewater treatment plants and land runoff. Several of these have been shown to have adverse effects on fish, including disruption of reproduction, growth and brain development. Previous studies have mainly focused on single compound exposures or simple mixtures and further evaluation of complex mixtures at low concentrations is needed. Within the framework of the European Horizon2020 project EDC-MixRisk, EDCs linked to adverse effects in neurodevelopment and growth in a pregnancy cohort study have been identified. Mixtures of these chemicals (phthalate metabolites, phenols and PFASs) were synthesized and subjected to be tested in a range of in vivo and in vitro systems. The thyroid hormones (THs) are one of the targets of interest as they are essential for brain development and disruption of this axis may lead to alteration of neurodevelopment. The current study aimed to determine the effects of the EDC-mixtures on larval behaviour and to identify disruption of TH-related gene expression in zebrafish (Danio rerio) during early development. Zebrafish embryos were exposed to Mix N and Mix G (mixtures correlated with adverse effects on neurodevelopment or growth in the epidemiological study) for 48h in concentrations equivalent to 0.01x – 100x human levels. Alterations of larval behavior caused by the exposures were studied as an endpoint for neurodevelopment since behavior integrates many biochemical processes and can be altered by endocrine disrupting chemicals. Larval locomotion was tracked using the ViewPoint ZebraBox and a protocol of alternating dark/light cycles. Quantitative PCR was used to determine the effects of the EDC mixture on the expression of thyroid related genes. Our results show that acute exposure to the mixtures significantly alter larval locomotion and expression of genes involved in TH signaling, including thyroid hormone receptors 

TH056 Contaminants of emerging concern in the North American Great Lakes: Assessing environmental mixtures in multigenerational exposure studies
N. Cipoletti, St. Cloud State University / Aquatic Toxicology Laboratory; L. Wang, St. Cloud State University; H.L. Schoenfuss, St. Cloud State University / Aquatic Toxicology Laboratory

In aquatic ecosystems such as the North American Great Lakes watershed, organisms are exposed to complex chemical mixtures throughout life, producing effects not anticipated in laboratory settings designed to test acute effects of single chemicals. By exposing fathead minnows through three generations, we aim to capture exposure effects during sensitive life stages. Through two separate multigenerational studies, we analyzed the effects of both urban and agricultural co-occurring contaminants at environmentally relevant concentrations in the Great Lakes watershed. Fathead minnows were housed in a flow-through exposure system and propagated for three generations (approximately one year of continuous exposure). Larval fish were analyzed for predator avoidance performance, feeding efficiency, and growth. Adult fish were analyzed for fecundity, biological indices, and racial and sex-related hormone pathways. Two urban and agricultural exposures resulted in growth alterations between treatments most likely due to density-dependent growth. Urban exposure indicated higher fecundity (both first and second generation) at low and environmentally relevant concentrations as compared to control and high treatments, potentially as a therapeutic hazard associated with the estrogenic nature of the mixture. Agricultural exposure indicated higher fecundity (both first and second generation) at low and environmentally relevant concentrations in the Great Lakes watershed. To risk assess the potential for first generation only, whereas the second generation demonstrated decreases in environmentally relevant concentrations as compared to low and control treated fish, highlighting the differences between adult only exposed fish (first generation) and lifetime exposure (second generation). Urban exposure demonstrated no changes to male plasma VTG (egg-yolk precursor protein), but agricultural exposure demonstrated increases in high exposed male fish (first and second generation) and environmentally relevant exposed male fish (second generation) due to the estrogenic nature of the mixture and exposure during different developmental stages between generations. Results indicate that mixtures, environmentally relevant in composition and concentrations, have the potential to alter growth, lead to reductions in fecundity, and elevate egg-yolk precursor proteins in male fish. Potential, yet unknown, consequences to the population level of exposed aquatic organisms may exist and warrant further study.

TH057 Contaminants of emerging concern in the North American Great Lakes: Assessing species sensitivity to environmental mixtures
S. Kohn, N. Ost, St. Cloud State University / Aquatic Toxicology Laboratory; N. Cipoletti, St. Cloud State University / Aquatic Toxicology Laboratory; L. Wang, St. Cloud State University; U. Hashay, H.L. Schoenfuss, St. Cloud State University / Aquatic Toxicology Laboratory

Contaminants of emerging concerns (CECs) have been detected ubiquitously in aquatic environments, and their endocrine-disrupting effects concern us. We identified typical CECs in rivers associated with agricultural and urban land use in the North American Great Lakes watershed. A mixture of agricultural CECs (AG) was assembled to mimic the environmentally occurring compounds and consisted of 8 chemicals, while an urban CEC mixture (UB) contained 11 chemicals including the known estrogenic compounds, bisphenol-A, estrone and nonylphenol. In addition to estrogenicities, CECs, Minnesota Pollution Control Agency reported that an exposure to CECs in the river water activated a peroxisome proliferator-activated receptor (PPAR)/retinoid X receptor (RXR) pathway in the transcriptome analysis of fathead minnow. Our laboratory found that an exposure to CECs induced a higher incidence of hepatic vacuolization in fathead minnow, which would be an obesogenic effect of CECs via a PPAR/RXR signaling pathway. Two isoforms of estrogen receptor (ESR) of fathead minnow, bluegill sunfish, American alligator or human was examined in the human embryonic kidney 293T cells by quantifying their transcriptional activities using estrogen-response elements and luciferase reporter gene in an exposure to CECs. PRARG/RXRA signals might not be involved in a CECs-exposure inducing a hepatic vacuolization in fathead minnow. This results indicate that efficacious disruptor activity exists in CECs, yet further investigation of PPAR/RXR signal are required in fathead minnow, PRARG/RXRA signals might not be involved in a CECs-exposure inducing a hepatic vacuolization in fathead minnow. These results indicate that efficacious receptors and species differ between CECs mixture, and further endocrine studies of CECs are required for a better understanding and prediction of CEC effects by utilizing a variety of receptors cloned from diverse threatened and endangered species.
Contaminants of Emerging Concern in the North American Great Lakes: Effects from simple exposures to complex mixtures
U. Hasbay, H.L. Schoenfuss, St. Cloud State University / Aquatic Toxicology Laboratory

Contaminants of emerging concern (CEC), including pharmaceuticals, personal care products and industrial agents may impact aquatic life. Previous studies have documented endocrine disruption, growth impairment and behavioral effects of CECs in some organisms. However, these studies did not consider the complex mixtures of CEC present in the environment. The purpose of the current study was to understand the impact of CECs in the environment. We tested the hypothesis that as the CECs in exposures increase, the apical endpoints will show significant differences from simple exposure endpoints. We assessed the potential of 21 commonly detected CECs on three life stages fathead minnows: embryo (developmental abnormalities, transcriptomics), juvenile (survival, escape performance, feeding efficiency, qPCR) and adult (survival, secondary sex characteristics, nest defense, courtship, boldness, qPCR) after 96-hour flow-through exposures. In addition, we began the process of building a series of complex mixtures to study the CEC effects using neural network methodology. Individual compound concentrations and mixture composition were based on an analysis of nearly 500 water samples collected as part of the Great Lakes Restoration Initiative. Our current findings show that the larval survival was significantly reduced (p<0.05, ANOVA) by diverse CECs including estrone, desvenlafaxine, and tris(2-butoxyethyl) phosphate exposures. Interestingly, exposure to ibuprofen showed a potential therapeutic effect at the medium concentration to assess two divergent treatment technologies of wastewater treatment and also indicated observation of endocrine disruption on our mixture exposure which includes all studied chemicals. We expect that these evaluations will lead us to improve adverse outcome pathway concepts by testing same chemical effects at different life stages of fathead minnows, and forming a linkage between behavioral responses and adverse outcomes.

TH061 Towards a multiparameter detection of biological effects caused by anthropogenic micro-pollutants
C.E. Reųänder, German Federal Institute of Hydrology; L. Moscovic, The Hebrew University of Jerusalem / Institute of Life Sciences, Department of Plant and Environmental Sciences; G. Reifferscheid, German Federal Institute of Hydrology / Department G Biochemistry Ecotoxicology

Organic micro-pollutants (MPs) can enter the aquatic environment via diverse pathways and sources e.g. waste water treatment effluents, agricultural activities or the disposal of various consumer goods. Tracking the occurrence, distribution and fate of MPs in the subsurface can provide insights into microbial activity of endocrine disruptors upon weathering or as a result of well bore leaching aquatic environments impact fish physiology and reproduction. Further research is underway to determine whether these observed effects have an impact at the population level.

TH062 Endocrine disruptors used in polymers in the offshore oil and gas industry
C. Phillips, University of Toronto; A. Smith, Cefas / Ecotoxicology

Endocrine disruptors used in polymers in the offshore oil and gas industry were identified and quantified in the offshore oil and gas industry. The study was conducted to assess the potential endocrine disruption of anthropogenic micro-pollutants. Laboratory cultured adult and larval fathead minnows were exposed to water samples from different sites in the two watersheds for 21 days. Adult minnows were analyzed for alterations in hematological characteristics (glucose, VTG, E2, 11-KT) and reproduction, while larval minnows were analyzed for feeding efficiency and predator avoidance performance. The Maumee River indicated reduced reproductive capability, as measured by fecundity, at specific sites where the channel width changed seasonal differences between the two sampling years potentially due to altered contaminant loads during heavier periods of precipitation. The Maumee River demonstrated no changes to larval predator avoidance behavior or other apical endpoints (feeding, growth) indicating that agricultural contaminants pose no/little perceived threat to larval development. Conversely, the Milwaukee River indicated increased reproductive capability, as fecundity increased among 2 of 7 sites in comparison to controls. Additionally, urban samples resulted in an increase in larval growth following 21 days of exposure, but did not impact the predator avoidance behavior. The results indicate that agricultural and urban pollutants entering aquatic environments impact fish physiology and reproduction. Further research is underway to determine whether these observed effects have a significant impact on the population level.
Thi063
Thyroid disorder screening using zebrafish as vertebrate model
I. Jurtíza, O. Jakab, C. Martí, A. Alzualde, BiOide; A. Muriana, BBD BioPhenix S.L./RD
Endocrine disrupting compounds are frequently found in the environment and have a profound impact on the development and physiology of vertebrate organisms. Thyroid Disrupting Compounds (TDC) specifically affects the function of thyroid hormones, interfering with their synthesis, transport and/or binding; altering important physiological processes. Several environmental contaminants such as polybrominated diphenyl ethers or halogenated organophosphates, used as plasticizer and flame retardant, are suspected to produce a thyroid-disrupting effect. Given so, chemical manufacturing entities could benefit from cost-effective methodologies for the screening of TDC in order to deselect candidates during the early phase of the development. In this work, we present an assay for the screening of potential TDC using zebrafish embryo. This vertebrate model is extensively used as a biosensor for the evaluation of acute and developmental toxicity, and several assays in zebrafish are described by the OECD guidelines for the testing of chemicals. Besides, the embryo’s small size and transparency allow to carry out fluorescence-based screening assays with medium throughput. In this work, the thyroid hormone disruption potential of several environmentally relevant contaminant was assessed. For this end, an initial acute toxicity assay was performed in order to estimate the EC50 and NOEC of the tested compounds, and subsequently select concentrations with no systemic toxicity. Afterward, change in the thyroglobulin (TG) synthesis was assessed using a zebrafish transgenic line expressing cyp19a1a, a marker of oogenesis in line with the 3R principles in relation to TG gene promoter, by analysis of the fluorescence microscopy images. Finally, a gene expression assay, using rt-qPCR, was performed over known markers of thyroid disruption to further characterize the involved pathways of endocrine disrupting effect. The zebrafish assay showed to be a sensitive and cost-effective assay to evaluate the potential thyroid disruptor activity of chemicals.

Thi064
Development of stably transfected cell lines with zebra fish thyroid hormone receptors alfa and beta for assessing endocrine disruption in environmental samples
V. Virgínia Herranz, INIA National Institute for Agricultural and Food Research and Technology; E. Sánchez Martínez, Instituto de Agua y Suelo S.A.; A. Alzualde, BiOide
The development of cell lines which express thyroid hormone receptors might provide a sensitive and rapid system to evaluate their potential effects. In addition, the use of mammalian cell lines has been considered as a key stepping stone for the establishment of new in vitro reporter gene assays with medium throughput. In this work, we describe the generation of HEK293 cells stably expressing human thyroid receptors α (hTRα) and β (hTRβ) together with luciferase gene (DR4-TK-Luc reporter construct) under the control of thyroid hormone response elements (TRE).

Thi065
Screening endocrine disrupting potentials of alternative plasticizers using thyroid hormone reporter assay
G. Lee, H. Kang, Seoul National University Graduate School of Public Health; K. Choi, Seoul National University / Environmental Health Sciences
Phthalates have been used as plasticizer in polyvinyl chloride (PVC), food containers, medical devices, building materials, and personal care products. Because of reproductive toxicity of several phthalates including bis(2-ethylhexyl) phthalate (DEHP) and diethyl phthalate (DEP), use of major phthalates are regulated in many products in several countries. Accordingly, many alternative plasticizers have been developed and increasingly used worldwide, but their possible adverse endocrine disruption effects are not well-known. The aim of this study is to screen endocrine disrupting potentials of several widely used alternative plasticizer, cyclohexane dicarboxylic acids (DINCH), acetyl tributyl citrate (ATBC), dioctyl terephthalate (DOP), trioctyl trimellitate (TOTM), bis(2-ethylhexyl) adipate, and dihexyl adipate (DEHA). A series of in vitro assays employing a human breast (MVLN), a human adrenal (H295R), and a rat pituitary (GH3) cells, were employed. The test doses for each plasticizers applied were determined based on preliminary toxicity assays for each cell line. While none of alternative plasticizers showed significant estrogen binding affinity in MVLN cells, DINCH and DEHA exhibited significant increase in estradiol (E2) to testosterone (T) ratio in H295R cells. These results suggest that these plasticizers DINCH and DEHA cause increased estrogenicity through altering steroidogenic pathway, similar to DEHP. In GH3 cell line, hTb gene was significantly downregulated by exposure to TOTM, suggesting its thyroid disrupting potential through altering signaling pathway to thyroid gland. Our observation shows that DINCH, DEHA and TOTM may disturb balance of important hormones. Further investigations using in vivo models are warranted.

Thi066
Development of reporter gene system for assessing cherry shrimp ecdysone receptor agonists using mammalian cells
K. Chan, The Chinese University of Hong Kong / Life Sciences; Y. Chan, K. Chu, The Chinese University of Hong Kong / School of Life Science
Ecdysoid is a key steroid hormone that regulates growth, development and molting in animals under the phyllum of Arthropod, which includes the insects and crustaceans. The hormone targets the receptor complex which initiates the ecdysone receptor (EcR) and retinoid X receptor (RXR). The activated complex anchoring on the ecdysone responsive element (ERE) stated on the promoter subsequently initiates transcription of the responsive gene(s). Chemicals act as receptor agonists do not necessarily adopt the structure of the native hormone, as in the case of estrogenic endocrine disruptors. Recently, for insect pest control, synthetic estriol hydrazides, DAH (α, β, γ, β, α, γ, β)-bis(2-ethylhexyl) adipate, and diethylhexyl adipate (DEHA). A series of in vitro assays employing a human breast (MVLN), a human adrenal (H295R), and a rat pituitary (GH3) cells, were applied. The test doses for each plasticizers applied were determined based on preliminary toxicity assays for each cell line. While none of alternative plasticizers showed significant estrogen binding affinity in MVLN cells, DINCH and DEHA exhibited significant increase in estradiol (E2) to testosteron (T) ratio in H295R cells. These results suggest that these plasticizers DINCH and DEHA cause increased estrogenicity through altering steroidogenic pathway, similar to DEHP. In GH3 cell line, hTb gene was significantly downregulated by exposure to TOTM, suggesting its thyroid disrupting potential through altering signaling pathway to thyroid gland. Our observation shows that DINCH, DEHA and TOTM may disturb balance of important hormones. Further investigations using in vivo models are warranted.

Thi067
Micro-injection as an alternative for aquatic exposure? A case study in zebrafish embryos with 17α-ethinylestradiol.
E. Meurich, University of Antwerp / Zebrafishlab Dept Veterinary Sciences; F. Lai, University of Antwerp / Toxicological Centre Dep of Pharmaceutical Sciences; L. Vergauwen, University of Antwerp / Zebrafishlab Dept Veterinary Sciences SPHERE; A. Covaci, University of Antwerp, Toxicological Center / Toxicological Centre Dep of Pharmaceutical Sciences; A.L. van Nuijs, University of Antwerp / Toxicological Centre Dep of Pharmaceutical Sciences; S.J. Van Cruchten, University of Antwerp / Applied Veterinary Morphology, Dep Veterinary Sciences; D. Knapen, University of Antwerp / Zebrafishlab Dept Veterinary Sciences
Pharmaceutical companies have to perform an environmental risk assessment for every drug that is launched to the market. The mandatory tests for potential endocrine disrupting potential through altering signaling pathway to thyroid gland. Our observation shows that DINCH degradation potential of several environmentally relevant contaminant was assessed. For this end, an initial acute toxicity assay was performed in order to estimate the EC50 and NOEC of the tested compounds, and subsequently select concentrations with no systemic toxicity. Afterward, change in the thyroglobulin (TG) synthesis was assessed using a zebrafish transgenic line expressing cyp19a1a, a marker of oogenesis in line with the 3R principles in relation to TG gene promoter, by analysis of the fluorescence microscopy images. Finally, a gene expression assay, using rt-qPCR, was performed over known markers of thyroid disruption to further characterize the involved pathways of endocrine disrupting effect. The zebrafish assay showed to be a sensitive and cost-effective assay to evaluate the potential thyroid disruptor activity of chemicals.
was different between both exposure routes, while vitellogenin (vtg) and estrogen receptor 1 mRNA levels were similar between both routes after EE2 exposure. At the morphological and physiological level we observed differences as well. However, the degree of ER-binging was similar between both routes from day 1 until day 5. Despite daily refreshment, the EE2 concentration in the medium decreased regardless of the exposure concentration. The internal doses were the highest at the beginning of the exposure for both exposure routes and decreased after 5 days. The order of magnitude of the internal dose was also similar between the injection and an aquatic exposure in the µg/L range, which was also seen e.g. for the mRNA expression of vtg. Based on the dose measurements we can conclude that even if the embryos were dosed with EE2 within the same order of magnitude that there were still different outcomes for some endpoints. Therefore micro-injection is rather a complementary method and not an alternative route for aquatic exposure.

**TH068**

Vitellogenin expression, ovarian growth and hormone levels are affected by atrazine in the crayfish Procambarus clarkii

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Atrazine, a widely used herbicide, has been categorized as a suspected endocrine disruptor for many years. Although several studies have investigated the effects of atrazine exposure on reproductive function, its safety remains controversial and the mechanisms of its toxicity remain unclear. In this study, we tested the hypothesis that atrazine can affect reproduction in crayfish through dysregulation of vitellogenin expression and hormonal synthesis. Adult female crayfish (*Procambarus clarkii*) were exposed during one month to atrazine at concentrations of 1 or 5 μg/L. At the end of the exposure, ovaries, hepatopancreas and hemolymph samples were harvested for analysis of vitellogenin expression and steroid hormone levels. Ovarian tissue was also sampled for both biochemical and histological analyses. Atrazine-exposed crayfish had a lower expression of vitellogenin in the ovary and hepatopancreas, as well as smaller oocytes and reduced vitellogenin content in the ovary. Despite these effects, circulating levels of estradiol increased in females exposed to 5 μg/L of atrazine, showing that the inhibiting effect of atrazine on vitellogenin production was not related to a lower secretion of sexual steroids; instead, some early stimulating effects of estradiol on vitellogenesis could have occurred. Furthermore, in the hepatopancreas, together, our data showed that atrazine exposure was able to inhibit vitellogenin production in the crayfish *P. clarkii*, altering on the other hand the normal balance of sex steroids.

**TH069**

Identification of molt-inhibiting hormone and ecdysteroid receptor sequences in Procambarus pulex and consequences of endocrine disruptor exposures

E. Gismundi, University of Liege

Endocrine disruptors (EDCs) are well known to disrupt the development and the reproduction of exposed organisms. Although this point has been studied in vertebrate models, the limited knowledge of the endocrine system of invertebrates makes the evaluation of EDCs effects difficult. However, invertebrates represent the other major part of the ecosystems, such as benthic organisms, which are crucial for their functioning (e.g. litter degradation, food resource). Moreover, gammarids are hosts of hidden parasites such as vertically transmitted microsporidia (*microsporidia*) which could be confounding factors in assessment of EDC effects, since microsporidia VT could feminize juvenile males in some *Gammarus* sp. Consequently, currently, no biomarkers (assessment tools) are available to assess the endocrine disruption in gammarids. The present work focused on EDC effects on the molt process of *Gammarus pulex*, by researching the DNA sequences of two main proteins in the endocrine system of amphipods: the molt-inhibiting hormone (MIH) and the ecdysteroid receptor (EcR). Next, the expression variations of these two genes have been measured by RT-qPCR after an exposure of four weeks to atrazine (4HT). Together, our data showed that in the hepatopancreas, as well as smaller oocytes and reduced vitellogenin content in the ovary. Despite these effects, circulating levels of estradiol increased in females exposed to 5 μg/L of atrazine, showing that the inhibiting effect of atrazine on vitellogenin production was not related to a lower secretion of sexual steroids; instead, some early stimulating effects of estradiol on vitellogenesis could have occurred. Furthermore, in the hepatopancreas, together, our data showed that atrazine exposure was able to inhibit vitellogenin production in the crayfish *P. clarkii*, altering on the other hand the normal balance of sex steroids. Next, the expression variations of these two genes have been measured by RT-qPCR after an exposure of four weeks to atrazine (4HT). Together, our data showed that atrazine exposure was able to inhibit vitellogenin production in the crayfish *P. clarkii*, altering on the other hand the normal balance of sex steroids.

**TH070**

Use of in vivo and in vitro assays to investigate the effects and bioavailability of endocrine disrupting compounds in sediment on the benthic invertebrate Chironomus riparius

S. Karnatz, RWTH Aachen University / Institute for Environmental Research Department of Environmental Analysis; S.E. Crawford, RWTH Aachen University / Institute for Environmental Research, Dept. of Environmental Analysis; V. Esser, RWTH Aachen University / Physical Geography and Geocology; A. Müller, RWTH Aachen University / Institute for Environmental Research; A. Shulaikevich, Institute for Environmental Research (RWTH Aachen University) / Institute for Environmental Research; H. Hollett, RWTH Aachen University / Institute for Environmental Research

Sediments act as a sink and source of chemicals in the environment and, therefore, it is of great importance to know how sediment-bound chemicals affect aquatic organisms. The synthetic hormone 17α-Ethynylestradiol (EE2), a component of oral contraceptives, is ubiquitous in the environment and is a known potent endocrine disrupting compound (EDC) that adversely affects aquatic vertebrates (e.g. reproduction and development, uptake of potential of sediment-bound EE2 on benthic organisms is important for understanding the potential effects on vertebrate predators and subsequent upper trophic levels as a secondary source of contamination.

**TH071**

Assessing acute toxicity of Bisphenol A on Daphnia magna by passive dosing approach

H. Kwon, Y. Jeong, H. Jeon, S. Kim, KIST Europe / Environmental Safety Group

Bisphenol A (BPA) is a raw material for widely used polycarbonate plastics, but it is known to have negative effects on human health and the environment. It was classified as an endocrine disrupting chemical (EDC), which requires an accurate and reliable toxicity data for robust risk assessment. However, currently available in vivo assays cannot be used as an exposure concentration (EC) that adversely effects aquatic vertebrates (e.g. reproduction and development, uptake of EE2 in daphnids at 10 mg/L). Through the passive dosing method, we were able to determine the sili-bound EE2 on benthic organisms is important for understanding the potential effects on vertebrate predators and subsequent upper trophic levels as a secondary source of contamination.

**TH072**

Conventional spiking method often fails to control the exposure concentration of *Daphnia magna* by passive dosing approach

E. Gismundi, University of Liege

Endocrine disruptors (EDCs) are well known to disrupt the development and the reproduction of exposed organisms. Although this point has been studied in vertebrate models, the limited knowledge of the endocrine system of invertebrates makes the evaluation of EDCs effects difficult. However, invertebrates represent the other major part of the ecosystems, such as benthic organisms, which are crucial for their functioning (e.g. litter degradation, food resource). Moreover, gammarids are hosts of hidden parasites such as vertically transmitted microsporidia (*microsporidia*) which could be confounding factors in assessment of EDC effects, since microsporidia VT could feminize juvenile males in some *Gammarus* sp. Consequently, currently, no biomarkers (assessment tools) are available to assess the endocrine disruption in gammarids. The present work focused on EDC effects on the molt process of *Gammarus pulex*, by researching the DNA sequences of two main proteins in the endocrine system of amphipods: the molt-inhibiting hormone (MIH) and the ecdysteroid receptor (EcR). Next, the expression variations of these two genes have been measured by RT-qPCR after an exposure of four weeks to atrazine (4HT). Together, our data showed that atrazine exposure was able to inhibit vitellogenin production in the crayfish *P. clarkii*, altering on the other hand the normal balance of sex steroids. Next, the expression variations of these two genes have been measured by RT-qPCR after an exposure of four weeks to atrazine (4HT). Together, our data showed that atrazine exposure was able to inhibit vitellogenin production in the crayfish *P. clarkii*, altering on the other hand the normal balance of sex steroids.
Toxic effects of juvenile hormone analogue insecticides, methoprene and fenoxycarb, on cherry shrimp (Neocaridina davidi) X. HU, The Chinese University of Hong Kong; K. Chan, The Chinese University of Hong Kong / Life Sciences; K. Chu, The Chinese University of Hong Kong / School of Life Sciences.

Crustaceans are a large group of arthropod, and they are the major constituents to the aquatic ecosystem that provide a variety of ecological and economic services. Nevertheless, the increasing quantities of insecticides leached into water bodies severely affect the health of aquatic environment globally and heighten the adverse impact on the crustaceans. Among these insecticides, juvenile hormone analogue (JHA) insecticides are a kind of endocrine disrupters known to interfere with the normal hormone action in insects by mimicking the juvenile hormones. However, the structure and functions of the methyl farnesate (MF) in crustaceans are similar to the juvenile hormone (JH) in insects. Therefore, the exogenous JHA insecticides would cause adverse effects on the development and reproduction in crustaceans as in insects. The aim of our study is to examine the toxic effects of JHA insecticides - methoprene and fenoxycarb on a freshwater shrimp Neocaridina davidi which is successfully cultured and maintained in our laboratory as a new crustacean model. These insecticides are growing in use in agriculture both locally and globally, and their impact to the aquatic ecosystem is needed to be further clarified. In the present study, the acute and chronic toxicity effects of two juvenile hormone analogue insecticides, fenoxycarb and methoprene, on newborn N. davidi were investigated. The 24h and 48h median lethal concentrations (LC50) of fenoxycarb and methoprene were 1.40, 0.97 μg/mL (4.64, 3.20 mmol/L) and 1.96, 1.26 μg/mL (6.32, 4.06 mmol/L), respectively. The results indicate the interference of these two JHA insecticides on the juvenile hormone system.

TH073 Development of Multimedia Fate Model for Human Risk Assessment of EDCs in the Asan Lake Watershed, Korea M. Choi, J. Kim, Greenecos Inc.; Y. Kim, Greenecos Inc.; CEO Molecular fate models (multimedia fate model for H/man Risk Assessment in Multimedia Environment) has been developed considering topographic features and environmental and been meteorological data of watershed region of Asan lake in Korea. Human risk assessment system using this multimedia fate model was established and integrated to assess spatial exposure and risk about human living in this area. For this purpose, first, GIS database system containing of environmental and meteorological data of watershed region was developed. Second, Environmental concentrations of various chemicals were predicted, applying different fate processes according to different chemical properties. Third, advection and dispersion by wind in air grids, runoff in watershed, flows of water in water segments are considered in the watershed-based multimedia fate model, which was linked to a risk assessment system. Fourth, exposure indicators were developed and multimedia fate model (Endocrine Disrupting Chemicals) and assess human risk in this area. HURAME is valuable tools for predicting the fate of chemicals in evaluative and real environments with areas consisting of many watersheds. These models are an integral component of exposure assessment and risk assessment strategies, and are used in detailed assessments of contaminant fate. The aim of these models is to describe quantitatively contaminant fate and migration in a defined watershed region, with water segments and air grids inside of the region of interest treated more complicatedly. As a result, regional levels of environmental contamination are controlled by environmental parameters and processes and meaningful evaluation requires assessment of contaminant fate in neighboring regions. A linked set of regional models may be the potential to describe quantitatively the impact of chemical emissions over a wider geographic scale with significant spatial differences in environmental characteristics and chemical use patterns.

TH074 Comparative toxicity and endocrine disruption potential of urban and rural ambient organic PM from the Asan River, G-3 cells. B.L. van Drooge, IDAEA-CSIC / Department of Environmental Chemistry; A. Marqueno Bassols, Institute of Environmental Assessment and Water Research (IDAEA-CSIC) B. Pina, C. Porte, IDAEA-CSIC / Department of Environmental Chemistry; J. Grimalt, Institute of Environmental Assessment and Water Research (IDAEA-CSIC) / Department of Environmental Chemistry.

Outdoor ambient air particulate matter and air pollution are related adverse effects on human health. The present study assesses the cytotoxicity and ability to disrupt aromatase activity of organic PM, extracts from rural and urban areas at equivalent air volumes from 2 to 30 m³, in human placental JEG-3 cells. Samples were chemically analysed for particle bounded organic compounds with endocrine disrupting potential, i.e. PAH, O-PAH, phthalate esters, but also for organic molecular tracer compounds for the emission source identification. Rural samples collected in winter were cytotoxic at the highest concentration tested and strongly inhibited aromatase activity in JEG-3 cells. No cytotoxicity was detected in summer samples from the rural site and the urban samples, while aromatase activity was moderately inhibited in these samples. In the urban area, the street site samples, collected close to intensive traffic, showed stronger inhibition of aromatase activity than the samples simultaneously collected at a roof site, 50 m above ground level. The cytotoxicity and endocrine disruption potential of the samples were linked to combustion products, i.e. PAH and O-PAH, especially from biomass burning in the rural site in winter. Exposure of zebrafish embryos to the same organic PM extracts from the rural site at the same concentrations led to weaker inhibition of aromatase activity and an induction of genes implicated in basic cellular functions, such as cell proliferation. Moreover, the embryo transcript analysis showed strong correlations between the Aryl Hydrocarbon Receptor signalling pathway and PAH concentrations. On the other hand, in the zebrafish embryos exposure experiment, the urban samples showed an induction of oxidative stress related genes, which suggest different potential adverse outcomes for exposure to air pollution from specific sources.

TH075 Dietary and non-dietary prenatal exposure to endocrine disruptors (BPA and DEHP), Spanish case study.

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Endocrine disruptors (EDs) are chemicals compounds that send confusing messages to developing organisms causing various disorders by mimicking or altering the effect of hormones in the human body. Several EDs such as Bisphenol A (BPA) and di(2-ethylhexyl)phthalate (DEHP), are involved in obesity and diabetes diseases in children. Recent studies have shown evidences that these chemicals can cross the placental barrier making fetal exposure closely related to maternal exposure. The aim of this research is to establish fetus exposure to EDs (BPA and DEHP). To address this issue, recently, pregnant women recruitment has begun. In present work, dietary and non-dietary (dermal, non-dietary ingestion and inhalation) exposure of these women was considered in order to predict the dose of EDs at which the child has been exposed in the early stage of the development. The data obtained from this cohort (such as, physiological data, dietary habits and lifestyle, among others) was implemented into a physiological based pharmacokinetic (PBPK) model, which assesses the absorption, distribution, metabolism and excretion of a chemical compound into human body, as well as the internal exposure to target organs. To estimate the early exposure of the child, the model was implemented with fetus compartment for these chemicals. Results indicates that for both, BPA and DEHP, diet is the main contributor to the total exposure. However, indoor and outdoor environment and dermal exposure also contribute significantly to the total DEHP exposure. Levels of both EDs were modelled in maternal blood and in fetus blood as well as in other body compartment. These results will be validated with the results of biological monitoring in the current cohort (n=72). The integration of the data obtained from current on-going human biomonitoring campaign and the physiological based pharmacokinetic model, here implemented; predict the early exposure of the child/fetus to EDs. This work is included in the frame of HEALS project (FP7-603946).

TH076 Sensitive Biomarker Assay using LC-MS/MS: Determination of Thyroid Hormones (T3 and T4) in Fetus, Pup and Adult Rat Serum - Sampling Considerations S. Diaram, Enviso / Bioanalysis (LC-MS/MS)

The regulatory need for the analysis of circulating levels of thyroid hormones in rats (fetus, pup and adult) on reproductive toxicology studies has prompted the need for a sensitive assay for T3 and T4, which could not be fulfilled when using the traditional radioligand assay format. A new method was developed with the use of tandem liquid chromatography coupled to tandem mass spectrometry detector. The multiplex method utilizes a 50 μL sample volume of serum to determine both T3 and T4 from the same sample aliquot. Across several studies from various Toxicology facilities was observed that there is a particular prevalent in fetus and Day 4 of age pups. Considering that samples are collected from animals of fetus and Day 4 of age pups, which may be triggered for analysis subsequent to Day 13 and adult male samples being analyzed, the emphasis of the integrity of the sample is paramount to the success of the analysis.
ensure meaningful data can be collected. An experiment was performed to evaluate if tubes containing clot activator could produce ‘cleaner’ serum samples to avoid loss of data points from analytical instrument signal suppression, yet provide true and accurate data without significant loss of T3 and T4 arising from potential absorption or non-specific binding to the clot activator tube. The CV (precision) and RE (accuracy) for both T3 and T4, across quality control samples (generated from collection tubes (a) and (b)) were within acceptance criteria of ≤20% (25% for the LLOQ) demonstrating that tubes containing clot activator can be used for T3 and T4 sampling.

TH077
Steroid estrogens and estrogenic activity analysis in dairy farm watersheds regardless of effluent management practices
L.A. Tremblay, Cathawron Institute; J.B. Gadd, NIWA / Department of Chemistry; G. Northcott, Northcott Research Consultants Limited
Steroid estrogens contamination has been linked to adverse effects on aquatic biota. Steroid estrogens are excreted by all mammals and are therefore found in most agricultural wastes including dairy manure and dairy shed effluent (DSE). Some previous studies have demonstrated elevated levels of free and conjugated estrogenic steroids in DSE and this source has increased as New Zealand has experienced rapid expansion and intensification of dairy farming. This research used an approach incorporating analytical chemistry and bioassays to evaluate the levels of estrogenic activity in environmental samples from representative dairy watersheds with differing DSE management practices: either auto-cstr or spread to streams. The obtained results suggest that direct access of cattle to streams, the direct input of DSE into waterways and runoff from land application all require more intervention and effective management.

TH078
Toxic receipt: Why You Should Avoid it?
J. Mila, Institute of Chemistry, Technology & Metallurgy / Department of Chemistry; V. Mart, J. Randjelovic, L. Šojić, ALHem - Safer Chemicals Alternative
Bisphenol A (BPA) is at the moment one of most commercial chemicals at global scale and is used in production of thermal papers as a color developer. BPA is not chemically bound to paper, so in contact with skin, it migrates into the skin, and is absorbed. This chemical is proved to be toxic for fertility, disruptive for endocrine activity and steroid estrogens were prevalent in the waterways within all the studied dairy watersheds. Estrone was the predominant steroid measured in watershed waters because of its presence in dairy cow wastes and as a degrade of the main dairy cow estrogen, 17α-estradiol. Measurable estrogenic activity (17β-estradiol equivalents) was found at low levels in 83% of the stream samples (highest: 1.44 ng L\(^{-1}\) - Eeq) and at the groundwaters (≤0.15 ng L\(^{-1}\) - Eeq). While estrogenic activity was generally -1, one of (10) stream with measurable estrone, 17α- and 17β-estradiol had activity of 1.4 ng L\(^{-1}\), a level potentially harmful to aquatic biota. Comparable steroid concentrations and estrogenic activity were found whether DSE was spray irrigated on farm paddocks or directly discharged into waterways. This suggests that direct access of cattle to streams, the direct input of DSE into waterways and runoff from land application all require more intervention and effective management.

Risk assessment of Nanomaterials: innovative approaches and application of recent research developments to regulatory science (P)

TH080
Evaluate the ecological risk during product development: safe by design case study - Met@link project
R. Baun, Technical University of Denmark / DTU Environment; N.B. Hartmann, Technical University of Denmark (DTU) / DTU Environment; A. Hager, DTU - VITO / ABS; B. Geerts, VITO NV / Health; s. verstraeten, VITO / ABS
Safe-by-design requires risk evaluation at critical points during the development of a product to enable a well documented choice for the lowest risk option. In the Met@link project new metal-based inks - enriched with Ag-nanoparticles (Ag-NP) - for printing conductive tags are developed. The environmental risk is assessed to support decisions between different environmental risk options (or risk management options). Ecotoxicological ecotoxicity testing is used to assess the potential effects on the one hand (concentration effect relations for the target organisms) and evaluates the potential exposure of target organisms on the other hand (i.e. to define the environmental compartments and organisms of concern). Risk management either reduces the potential effects (f.i. redesigning the product) or prevents the predicted exposure (f.i. redesign the production process) to minimize the potential risk. Case study: ERA Ag-NP metal based ink. Potential effects of concern: Literature data learn that Ag-NP particles are indeed highly toxic to aquatic ecosystems, mainly due to the leaching of Ag+ ions. Potential exposure? Looking at the production process the aquatic ecosystem is of concern because of the waste water generated during production of the inks. Two prototype inks with suitable technical properties were formulated. Both were tested for their leaching potential and for their ecotoxicity to aquatic organisms. Ag-concentrations (ICP-AES) and Ag-NP (SP-ICP-MS) were measured, and the ecotoxic effects on algae (OECD 201) and Daphnia (OECD 202) were measured. Results showed that proper coating material helped to prevent leaching of Ag+ and substantially decreased the ecotoxicity of the leaching fraction.

TH081
REACH Substance Evaluation of silver - justification of read-across from ionic silver to nanosilver
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As part of the REACH Substance Evaluation for silver, new data was required to be generated and further justify the read-across from ionic silver to silver nanof orm. Information on aquatic and soil ecotoxicity of the smallest silver nanof orm with the highest specific surface area registered under REACH as compared to ionic silver. The tested nanof orm shall be sufficiently characterised; Information on the fate of nanosilver in soil only in case any of the ecotoxicity tests show higher toxicity for nanosilver as compared to ionic silver; Information on the uses for each individual nanof orm registered under REACH. An ecotoxicity testing programme was therefore undertaken comparing the effects of nanosilver with silver nitrate using 3 internationally standardised and accepted aquatic ecotoxicity tests: Toxicity to the alga Pseudokirchneriella subcapitata (OECD Test Guideline No. 201); nanosilver was less toxic than silver nitrate. Long-term toxicity to Daphnia magna (OECD Test Guideline No. 211): nanosilver was less toxic than silver nitrate. Toxicity to soil microorganisms (OECD Test Guideline No. 216) in 3 soils representative for the EU: nanosilver was equally or less toxic than silver nitrate. The silver nanof orm was fully characterised (aqueous suspension containing approximately 37% nanoparticles with spheroidal-like shape, mean primary particle size 9.4 nm). The dissolution behaviour of the tested silver nanof orm was determined in the test methods used in the ecotoxicity tests. The nanosilver dissolution behaviour qualitatively explained the observed toxicity. Since the ecotoxicity testing demonstrated that nanosilver was equally or less toxic than ionic silver, further fate testing in soil was not required. The data collection on the uses of the silver nanof orms covered by the REACH registration dossier showed limited tonnage and ecotoxicity. Since the ecotoxicity testing demonstrated that nanosilver was equally or less toxic than ionic silver, further fate testing in soil was not required. The data collection on the uses of the silver nanof orms covered by the REACH registration dossier showed limited tonnage and use of nanosilver. Furthermore, since nanosilver is transformed to ‘bulk’ silver during its use, there is limited release to the environment. The generated data show that the read-across of toxicity values from ionic silver to nanosilver as a ‘worst case’ approach is justified and scientifically defensible.

TH082
Revising REACH technical guidance on information requirements and chemical safety assessment for engineered nanomaterials for aquatic ecotoxicity endpoints - recommendations from the EnvNano project
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This study revises the technical guidance on information requirements and chemical safety assessment for engineered nanomaterials for aquatic ecotoxicity endpoints - recommendations from the EnvNano project. The study evaluates the current technical guidance and highlights areas for improvement. The study further evaluates the current information requirements and chemical safety assessment for engineered nanomaterials for aquatic ecotoxicity endpoints. The study identifies gaps in the current technical guidance and recommends improvements. The study also evaluates the current information requirements and chemical safety assessment for engineered nanomaterials for aquatic ecotoxicity endpoints. The study identifies gaps in the current technical guidance and recommends improvements. The study also evaluates the current information requirements and chemical safety assessment for engineered nanomaterials for aquatic ecotoxicity endpoints.
Engineered nanomaterials (ENM) are used in different products with the consequence that they can be released into the environment during their life cycle. Given the large variety of ENMs, the effort for an individual investigation and assessment would be enormous. Therefore grouping of ENMs and read across between different materials is a major target for future risk assessment. In this poster we present practicable approaches that can support the discussion on grouping of ENMs regarding their environmental potential. In our project we focused on the behaviour of the pristine ENMs in aquatic and terrestrial compartments. The transformation (chemical transformation and dissolution) and the transport (mobility and agglomeration) of an ENM in the environment was studied. To predict the exposure potential for the environmental compartments both pieces of information were combined to result in a number (fate bond from 1 to 3 for high) for a so called “fate bond” which will be included in a matrix of ENM grouped regarding their potential environmental risk. For example, if the transformation via dissolution and chemical transformation is low in the environmental compartment, the transformation potential of the ENM is low. If the mobility is low and the agglomeration potential is high, the transport is also low. Low transformation and low transport means high ENM exposure potential in the compartment considered and leads to a number value of "3" in the fate bond. For simplification, in this project water phase and sediment phase are considered as one compartment (water compartment) and therefore transport and mobility affected by e.g. agglomeration and sedimentation are not needed to be considered in the presented approach. In contrast, for soil systems the mobility was analysed in detail, as important factor for the exposure concentration. For an environmental risk assessment the fate information is base (fate bond) is considered in the decision matrix on the production volume of the ENM, that portion which is relevant for the considered use, used in closed/open systems, and slow/fast release into the environment. The resulting so called “release bond” classes ENMs to ENMs with low release (release bond = 1) to high release (release bond = 3). The release grouping is followed by a detailed description of release scenarios for the considered use. These scenarios allow the identification and assessment of the potential environmental risk that the ENM is emitted. Furthermore, possible sinks become obvious for which the fate and ecotoxicological effect grouping need to be performed. Basically, surface water, sediment and soil are possible sinks in the environment. To simplify the fate grouping approach, surface water plus sediment are here supposed to be a monophase system. Thus, chemical transformation and dissolution are considered as relevant processes for the aquatic fate grouping. Transformation (chemical and dissolution) and transport (agglomeration and movement) are considered relevant processes for the terrestrial fate grouping. The so called “fate bond” classes ENMs to ENMs with low exposure potential (fate bond = 1) to high exposure potential (fate bond = 3). In a next step, release bond and fate bond are combined to a so called “ecotox bond” (1 to 9). The ecotox bond is used for the so called ecotox bond. The latter one is based on information about ecotoxicity of the bulk material, morphology of ENM, and the ion release potential. The combination results in a 5 x 5 risk matrix with 25 possible combinations of exposure and ecotox bonds. These are summarized to three risk groups low, medium and high. The applicability of the approach will be demonstrated by risk grouping of nano-ZrO and nano-TiO used in sunscreen products. Key words: release, fate, ecotox bond
The grouping of engineered nanomaterials (ENMs) is being intensively discussed in order to develop approaches that allow an adequate hazard assessment of ENMs while reducing the testing effort or to rank them regarding their environmental hazard. Two approaches differing in their focus have been developed and evaluated with a set of 25 ENMs. Based on systematic testing using aquatic test designs used in regulatory testing, the physical-chemical (PC) properties, i.e. morphology and reactivity, as well as ecotoxicity of the ENMs were identified as relevant parameters. The zeta-potential was considered to be less important. Regarding the parameters ecotoxicity of the bulk chemical, solubility and reactivity we decided upon a pragmatic approach with questions which have to be answered “yes” or “no”. Approach I (ecotox flow-chart) is characterized by maximum 24 growth points. For the parameter “morphology” only fibers and small spherical ENMs are considered. In the ecotox-bond every “yes” for an answer gives one point. The points are added together resulting in five groups in a range or band of 1 to 5 which is used for further assessment. The same number of points can be achieved by different properties resulting in groups of ENMs which can differ significantly in their PC-properties. This procedure is considered suitable for the initial prediction of ENMs that are new or that have been subjected to an environmental fate. Both approaches are considered to be a suitable starting point for further discussions and developments. Besides the definition of threshold values for solubility, fiber morphology and size of small spherical ENMs, further parameters (e.g. attachment of ENMs to algae) have to be explored to improve the consistency of the groups. Regarding the terrestrial ecotoxicity, soil properties seem to reduce the impact of the toxic properties of the ENMs. Currently the prediction of terrestrial toxicity is not satisfactory.

**TH087**

Forms of released engineered nanomaterials: A systematic assessment in material flow analysis

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The forms in which engineered nanomaterials (ENMs) are released to the environment affect their fate and toxicity, two parameters essential to risk assessment. Yet, most of current models assessing ENM releases to the environment do not fully account for the transformations that they undergo before release to the environment. This work consists in the development of a method based on current literature, expert elicitation and probabilistic material flow analysis (PMFA) for modelling the proportions of nano-Ag and nano-TiO₂ flowing in different forms (e.g. dissolved, aggregated nanoforms and nanoparticles) in the major environmental compartments. The ENMs flow from production, manufacturing and use to wastewater treatment (sewer system, wastewater treatment plant and sludge) and solid waste treatment (landfilling, incineration and recycling) before reaching air, soil or surface water. Each mass flow was described with a probability distribution. The variability of the data obtained in the literature was used to assess the width of these distributions. Nano-Ag is released to surface water and soil mainly in transformed forms (61% and 77%, respectively), while nanoparticulate forms dominate the releases to air (60%). Most transformations occur in water. Nano-TiO₂ presents contrasting results, as most of the releases to air, soil and water are in nanoparticulate forms (80%, 94% and 99%, respectively). The only transformation identified in air is aggregation (86%, 90% and 99%, respectively). These results show that, especially for nano-Ag, the actual nanoparticulate form represents only a small fraction of the total ENM mass released to the environment, thereby calling for a revision of current exposure levels commonly used.

**TH088**

Using the SimpleBox4.0n tool for predicting the environmental concentration of nanomaterials

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In environmental risk assessment the risk quotient, predicted environmental concentration (PEC) relative to the predicted no effect concentration (PNEC), is a useful indicator for risk of chemicals. The SimpleBox4.0n modelling approach has long been applied in the regulatory framework REACH, as part of EUSES, to calculate PECs. The SimpleBox model was recently extended for use with nanomaterials (SimpleBox4.0-nano), by updating particle specific transport process algorithms and including nanomaterial specific transformation processes, such as agglomeration and dissolution. In this study we show the sensitivity of SimpleBox4.0-nano to the newly added process parameters. This shows that in addition to the dissolution rate, the attachment efficiency, and the concentration of natural particles and their size play a role. In order to use SimpleBox4.0-nano we provide guidance on measuring or calculating the relevant input parameters. Furthermore, we indicate the relevance of the different fractions of PECs as calculated by SimpleBox4-nano for estimating the risk quotient. 1. www.rivm.nl/simplebox4n

**TH089**

Directions of in silico method development to complement the predictive models used in risk assessment of nanomaterials

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There is an increasing need for predictive risk assessment of nanomaterials (NMs) that are new or that have been subjected to an environmental fate. Both approaches are considered to be a suitable starting point for further discussions and developments. Besides the definition of threshold values for solubility, fiber morphology and size of small spherical ENMs, further parameters (e.g. attachment of ENMs to algae) have to be explored to improve the consistency of the groups. Regarding the terrestrial ecotoxicity, soil properties seem to reduce the impact of the toxic properties of the ENMs. Currently the prediction of terrestrial toxicity is not satisfactory.

**TH090**

NanoScreen - Minimizing the risk associated with nanomaterials used in sunscreen at all lifecycle stages

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Among cosmetics and personal care products, sunscreen products are of emerging concern regarding both environmental and human health. While some organic UV blockers have been evidenced to undergo rapid photodegradation, to induce allergenic skin reactions due to dermal penetration, or to cause deleterious effects on marine system, the fate of mineral UV blockers is still under consideration from a regulatory perspective. This is largely related to the potential impact of nanotechnology-based products on both environment and human health. The nano-TiO₂ UV-blockers typically used in sunscreen usually consist of rutile nanoparticles coated with a first mineral layer of silica or alumina aimed at blocking the photocatalytic character, and thus passivating the nanomaterial. In addition, the grafting of a second layer of organic coating is aimed at favoring the nanomaterial dispersion. The fabrication and end of life steps are mainly considered and studied using methods that are rapid, accurate and resource efficient. To fulfill this need, we analyze a selection of currently available nanomaterials and NM-related parameters in exposure modelling seems eminent. In order to find the relevant application of new in silico methods, we analyze a selection of currently available and environmental risk assessment models for NMs. This analysis was done by identifying all the NM-related properties used in these models related to three categories of data: 1) Measured hazard or exposure data, 2) Extrinsic NM Properties, e.g. related to the interaction of the NM with the surrounding matrix, and 3) Intrinsic characteristics specific to the NM, matrix or experimental conditions. This analysis is combined with the current state of Quantitative Structure Property Relationships (QSPRs) development, as a specific set of in silico methods, to recommend further development of in silico methods for predictive risk assessment of NMs. In particular, the use of descriptors related to the interaction between a NM and its surroundings, e.g. the attachment efficiency, is proposed. QSPRs as well as other in silico methods are well suited to fill this gap in predictive risk assessment, under the condition that enough data of reliable quality becomes available.

**TH091**

Multimedia Modeling of Engineered Nanoparticles with SimpleBox4nano: Model Definition and Evaluation.

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Furthermore, we indicate the relevance of the different fractions of PECs as calculated by SimpleBox4-nano for estimating the risk quotient. 1. www.rivm.nl/simplebox4n

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SETAC Europe 28th Annual Meeting Abstract Book
A new test method to determine the bioaccumulation of manufactured nanomaterials in filtering organisms (Bivalvia) using the freshwater mussel Corbicula fluminea

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The identification and scientific assessment of compounds that bioaccumulate in organisms and biomagnify in food webs play a key role within the PBT-assessment. The bioaccumulation potential of compounds is commonly expressed in form of bioconcentration factors (BCF) determined in flow-through studies with fish according to OECD 305. Comparable studies with manufactured nanomaterials (MNMs) are difficult to carry out due to the lack of suitable test systems that allow a permanent and constant exposure of the compounds. MNMs tend to sediment in water and are supposed to be primarily taken up by benthic species in aquatic ecosystems. Different studies have shown that mussels are able to ingest and to incorporate MNMs suspended in water. However, existing standardised test methods to investigate the bioaccumulation of substances in mussels have been developed and optimized for soluble, non-particulate substances. Therefore, an alternative test concept was developed allowing to investigate the bioaccumulation of MNMs in bivalves under flow-through conditions. First studies were carried out with the freshwater mussel Corbicula fluminea. By using silver MNMs (NM300K) and silver nitrate we were able to compare the accumulation and elimination of ionic and nanoparticulate silver. Mussels were exposed for a period of 4 - 6 days. In both cases steady state concentrations of total silver in the mussel tissue were reached within 24 hours. The quantification of the silver in water was carried out by ICP-MS or ICP-OES. The dissolved and particulate silver concentrations were used to determine bioaccumulation factors for both test items. In a further study the bioaccumulation of a titanium dioxide nanomaterial (NM 105) was tested. The studies have shown that the new test system is suitable to investigate the bioaccumulation of MNMs.
The aim of this work is to investigate i) whether biochar can be used as a sorbent for metal contaminated soils and an activated biochar (aBC) (used for the PFAS study) for the removal of inorganic contaminants from soil. Biochar (BC) has a broad range of properties, such as heat, water and oil resistance. It is used for a variety of applications. However, in order to ensure a fully legitimate comparison between conventional (i.e. before oxidation) and after TO analyses, a number of critical knowledge gaps remain to be bridged. The two types of water samples (i.e. before TOP versus after TOP) might reveal differential instrument matrix effects or necessitate different clean-up strategies, which could de facto impact the method reporting limits and preclude a consistent comparison between the two approaches. The present work aimed at assessing the applicability of the TOP assay to various water matrices through stringent validation. The performance of a workflow involving persulfate oxidation followed by ultra-high performance liquid chromatography tandem mass spectrometry (TO-PHPLC-MS/MS) analysis was therefore evaluated using various environmental waters. The validation endpoints ascertainment included, notably, the evaluation of oxidation yields in the various matrices and TOP assay as a critical assessment of the toxicity effectiveness which occur at the instrumental analysis stage. The method was applied to a limited survey of surface water and groundwater samples. It was observed that even though fluorotelomer sulfonates (ZFTSAs) were the target pre-PFAs predominantly reported before oxidation in most instances, they could only partially account for the observed APFAA (molar concentration increases upon oxidation). The unexplained APFAA portion likely results from the oxidation of untargeted pre-PFAs for which oxidation yields are yet to be determined.

**TH096**

Use of biochars for the sorption of poly- and perfluorinated alkyl substances (PFAS) and heavy metals from contaminated soils

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The contamination of soil with a mixture of compounds represents a worldwide environmental issue. Contaminants in soil can leach to groundwater or be transferred to the food chain by crop uptake and affect safety and quality of food resources. Of particular concern is the contamination of soil with perfluorinated and/or polyfluorinated substances, which are ubiquitous in the environment and can occur in the environment.

The study aimed to investigate the effects of BC on the sorption of both organic and inorganic pollutants from soil. Biochar (BC) has a high adsorption potential for organic and inorganic contaminants and can be used at a low cost. Biochar is thus a promising and economic alternative to other carbonaceous materials, such as activated carbon, for this environmental application. In this study, three BCs were used as sorbents: a wood BC (wBC) made from wood chip waste (used for all the treated soils), an iron enriched BC (Fe-BC) (used for the metal contaminated soils) and an activated biochar (abC) (used for the PFAS contaminated soils). Isotherm batch tests have been carried out using a water and soil mixture (L/S=10) to which BC was added at increasing doses (from 0 to 20%). The aim of this work is to investigate i) whether biochar can be used as a sorbent for metal contaminated soils, ii) whether biochar can sorb PFAS in soils with both high and low TOC contents, iii) whether iron enriched BC increases the sorption of metal as compared to non-enriched BC and iv) whether there is a correlation of BC properties (surface area, pores, surface property, etc) with sorption.

**TH097**

Sorption of 14 PFAS to organic soil constituents - the effect of H+ , Na+, Ca2+ and Al3+ ions

H.F. de Campos Pereira, Swedish University of Agricultural Science / Department of Soil and Environment; M. Ullberg, Swedish University of Agricultural Sciences / Department of Soil and Environment; D. Berggren Kleja, Swedish University of Agricultural Sciences; I. Gustadsson, Swedish University of Agricultural Sciences / Department of Soil and Environment; L. Ahrens, Swedish University of Agricultural Sciences SLU / Dept. of Aquatic Sciences and Assessment Environmental Risk assessment of perfluoralkyl substances (PFASs) requires accurate prediction of their sorption in soils. The aim of this study was to investigate sorption of 14 PFASs, including perfluorocarboxylates (PFCAs), perfluorosulfonates (PFASs) and perfluorooctanesulfonamide (FOSA), to an organic soil horizon and the effect of solution pH and simulated soil organic matter (SOM) net charge as a function of pH and added concentrations of Al3+, Ca2+ and Na+. Generally, the organic C-normalized partitioning coefficients (Kc normalized) were nearly constant (1.32 ± 0.11 log unit (mol kg⁻¹ per pH unit) in H+ and 0.34 ± 0.40 log unit (mol kg⁻¹ per log unit) in Na+). The sorption increased with increasing perfluorocarbon chain length (hydrophobicity) for both PFBCs and PFASs with 0.60 and 0.83 log units per CF, respectively. Comparing the effect of the PFAS functional head group on sorption, affinity followed the order PFCSs < PFASs < FOSA. Effects from cation competition were small and instead sorption was more strongly related to the pH value. This suggests that the long-chained PFAs have a binding preference towards the highly condensed parts of the humic fraction of SOM, in similarity to other hydrophobic organic compounds, whereas shorter PFASs to a higher degree are bound to humic and fulvic acid where co-sorption of cations gives significant effects. A conceptual model which explains the observed difference in sorption behaviour between shorter and longer PFASs is presented. Progresses made on PFAS binding to organic soil fractions will contribute to more accurate prediction of PFAS sorption in soils and thereby aid in the environmental risk assessment of these chemicals.

**TH098**

Environmenal degradation rates for new PFAS via decarboxylation potential in water, in a MS-collision cell and dissolution

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Straight-chain perfluoroalkylphatic carboxylic acids, like PFOA, are extremely stable chemical compounds. In contrast, several other perfluorinated carboxylic acids are less stable and undergo decarboxylation - spontaneous degradation with loss of carbon dioxide. For instance, perfluorobenzoic acid decomposes slowly in aqueous solution, while perfluoropropionic acid decomposes so fast at room temperature that its spontaneous decomposition is a synthetic method for nonafluoroisobutane. There are indications that novel oxygen-containing analogs of PFOA are less stable towards decarboxylation. A typical detection method for PFACs is based on the same decarboxylation process: SRM transition from [M−1] to [M−45]. A collision energy, required for such transition is a measure of intrinsic stability of a compound. The competitive sorption of perfluorocarboxylic acids for this transformation can be satisfactorily predicted by DFT calculations at standard B3LYP/6-31+G(d,p) level. Decarboxylation rates in water for perfluorinated and structurally similar carboxylic acids also correlate well with MS and DFT-derived energies. Thus mass-spectral information and results of simple quantum-chemical modeling can be used as a measure of abiotic degradation potential for per- or perfluorinated acids in aquatic environment.

**TH099**

Perfluoralkylated acids (PFAAs) in soil and invertebrates (Isopoda) near a fluorocchemical plant in Flanders, Belgium.

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Perfluoralkylated acids (PFAAs) have been produced for over five decades. Due to their hydrophobic and lipophobic character they are suitable for a wide range of applications. However, PFAAs may enter the environment, accumulate in wildlife and pose a hazard and risk for human health and the ecosystems. The soils used in this work are contaminated with As, Ba, Cd, Cr, Cu, Hg, Mo, Ni, Pb, Sb, Se and Zn. Among the remediation techniques, the most common method is the combination of chemical and biological degradation of both organic and inorganic pollutants can be found. In the present study four soils with different types of pollution (poly- and perfluorinated alkyl substances (PFASs) and heavy metals) and two different total organic carbon contents (high and low), were used. PFASs are a class of compound characterised by hydrophobic, alkylated, fluorinated-saturated carbon-chain with a hydrophilic head attached at a terminal end. PFASs are hydrophobic and lipophobic and are widespread in the environment.

The aim of this study was to investigate sorption of 14 PFASs, including perfluorocarboxylates (PFCAs), perfluorosulfonates (PFASs) and perfluorooctanesulfonamide (FOSA), to an organic soil horizon and the effect of solution pH and simulated soil organic matter (SOM) net charge as a function of pH and added concentrations of Al3+, Ca2+ and Na+. Generally, the organic C-normalized partitioning coefficients (Kc normalized) were nearly constant (1.32 ± 0.11 log unit (mol kg⁻¹ per pH unit) in H+ and 0.34 ± 0.40 log unit (mol kg⁻¹ per log unit) in Na+). The sorption increased with increasing perfluorocarbon chain length (hydrophobicity) for both PFBCs and PFASs with 0.60 and 0.83 log units per CF, respectively. Comparing the effect of the PFAS functional head group on sorption, affinity followed the order PFCSs < PFASs < FOSA. Effects from cation competition were small and instead sorption was more strongly related to the pH value. This suggests that the long-chained PFAs have a binding preference towards the highly condensed parts of the humic fraction of SOM, in similarity to other hydrophobic organic compounds, whereas shorter PFASs to a higher degree are bound to humic and fulvic acid where co-sorption of cations gives significant effects. A conceptual model which explains the observed difference in sorption behaviour between shorter and longer PFASs is presented. Progresses made on PFAS binding to organic soil fractions will contribute to more accurate prediction of PFAS sorption in soils and thereby aid in the environmental risk assessment of these chemicals.
observed among the studied sites, but TOC was positively correlated with multiple PFAAs, including PFOS and PFHxS. This may indicate that TOC is an important factor influencing PFAA concentrations in coastal waters.
Oceanography
Fluorotelomer alcohols (FTOHs) and other poly- and per-fluorinated alkyl substances (PFASs) are common and ubiquitous by-products of various industrial telomerization processes. They can degrade into various perfluorinated carboxylic acids (PFCAs) including perfluorooctanoic acid (PFOA) and perfluorooctanesulfonic acid (PFOS), which are persistent organic contaminants of concern. This study assessed the use of polyethylene passive samplers as a sampling tool for volatile PFAS precursors coupled to their analysis via gas chromatography-mass spectrometry (GC/MS). Parallel active and passive sampling was also present in ambient air in Providence (RI USA) in April 2016. Atmospheric concentrations were dominated by FTOHs (average 9.9 – 16 pg/m3), with traces of other volatile PFASs also present. Polyethylene-air partitioning constants, log K_{pes}, were determined. A deployment at a Wastewater Treatment Plant (WWTP) was also performed in 2016, and sampling rates derived from the loss of performance reference compounds. Best-fit curves were used to determine polyethylene-air partitioning constants, log K_{pes}, during the 3-week uptake experiments. Derived log K_{pes} values for 6, 8, 12 and 10 FTOHs were 3.8, 4.4 and 4.8, respectively. For MePFOS and EtPFOS, derived log K_{pes} values were 4.0 and 4.4, respectively. Based on these partitioning constants, aqueous concentrations in the effluents were below 1 ng/L for the FTOHs, MePFOS and EtPFOS.

**TH105**

Occurrence and Removal of perfluoroalkyl and polyfluoroalkyl substances (PFASs) in full-scale water and wastewater treatment plants

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Perfluoroalkyl and polyfluoroalkyl substances (PFASs) comprise a group of compounds that are widely used in the markets for stain repellents, polishes, paints and coatings. In recent years, the occurrence of PFASs in the environment has been recognized as emerging environmental problem due to their persistence, bioaccumulability and potential bioactivity in organisms. Unlike most other persistent and bioaccumulative organic pollutants, PFASs are water soluble. Therefore, removal of PFASs by water treatment processes could be a challenge. The objective of this study was to evaluate the ability of different water treatment techniques to remove PFASs from water. In this study, three full-scale water treatment plants were investigated during a one-year monthly sampling for the removal of 31 PFASs, including 20 perfluoroalkyl acids (PFAAs) and 11 PFNA precursors. The treatment processes include conventional activated sludge trickling filters (CAS) and membrane bioreactor (MBR) system in plant 1 and 2, and microfiltration, reverse osmosis (RO), ultraviolet disinfection (UV) in plant 3. Short year monthly sampling for the 31 PFASs were performed in ambient air in Providence (RI, USA) in April 2016. Material and Methods A bioaccumulation monitoring of selected fish species (Hemiculter leuciscus, Leuciscus idus, Hydropsyche sp., Erpobdella octoculata and Leucophaea sp.) was performed in ambient air in Paris, France. A total of 16 PFAAs and their precursors were found in the samples of fish liver and juvenile fish exceeded EQS for PFOA (9.1 μg/L).

**TH107**

Analytical strategy to study the distribution of perfluoroalkyl substances in fish tissue of Italian deep subalpine lakes


Perfluoroalkyl substances, such as perfluorinated sulfonic acids (PFSAs) and perfluorinated carboxylic acids (PFCAs), are ubiquitous contaminants in the aquatic environment, including wildlife and humans. Perfluoroalkyl acids bind to proteins and the binding in bioaccumulation behaviour different from that of lipophilic substances. Therefore, conventional methods (fish fillet analysis, lipid normalization, etc.) to monitor their concentrations in aquatic biota cannot be used as such to assess the bioaccumulation and biomagnification of PFCA and PFSA. In this study, conventional monitoring approaches and new strategies are compared to assess the best methodology to be implemented in biota monitoring plans for these compounds. Several fish species from Lake Maggiore (Italy) were sampled to assess the occurrence in biota in the urban river Orge (near Paris, France). A total of 16 PFAAs and 10 of their potential precursors (pre-PFASs) such as 8:2 and 10:2 FTSA, as well as FOSA, N-MeFOSAA, N-EtFOSAA and 6:2 diPAP. However, a significant decrease in pre-PFCAs/PFCAs concentrations with trophic level suggested a possible contribution of

**TH108**

Potential contribution of targeted and unknown precursors to the apparent biomagnification of perfluoroalkyl acids (PFAA) in the food web of an urban river

C. Simonnet-Laprade, University of Bordeaux UMR EPPOC; H. Budzinski, University of Bordeaux; K. Maciejewski, UMR EPPOC; K. Le Menach, UMR CNRS EPPOC Universite Bordeaux / UMR UMR 5055; R. Santos, Hepia University of Applied Sciences Western Switzerland; F. Billet, A. Goutte, EPHE / UMR METIS; P. Labadie, UMR CNRS EPOC Universite Bordeaux / UMR UMR 5055

This study assessed the potential contribution of targeted and unknown fluorotelomer alkyl (FTAOs) or perfluoroalkyl acids (PFAAs) to biomagnification in caged fish fed commercial fish feed in the trophic web of the urban river Orge (near Paris, France). A total of 16 PFAAs and 10 of their potential precursors (pre-PFAs) including 4 perfluorooctane sulfonamide derivatives, 4 fluorotelomer sulfonates (FTSAs) and 2 polyfluoroalkyl phosphate (diPAPs) were analyzed in water, sediments and biota samples including invertebrate and fish species. PFASs were ubiquitous in all compartments and 22 mixed-effect models were detected in fish. PFAAs concentrations were from 0 to 147 ng·g⁻¹ wet weight and PFOS, PFPeDA and PFtDA were the dominant compounds. Pre-PFAs such as 6:2, 8:2 and 10:2 FTSAs, as well as FOSA, N-MeFOSAA, N-EtFOSAA and 6:2 diPAP were also frequently detected (60–100 %) and the sum of these compounds contributed to 1–18 % of 2PFAs. Trophic magnification factors (TMFs) were estimated using a generalized linear mixed-effect model and were > 1 for C1–C4 PFCAs and C2–C6 PFCA, as well as several pre-PFAs such as 8:2 and 10:2 FTSAs, FOSA, N-MeFOSAA, N-EtFOSAA and 6:2 diPAP. However, a significant decrease in pre-PFCAs/PFCAs concentrations with trophic level suggested a possible contribution of

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precursors to the apparent biomagnification of PFCAAs, via their biotransformation. In addition, the Total Oxidizable Precursor (TOP) assay was applied to sediments and, for the first time, to biota samples. Results revealed the presence of large proportions of unknown p-PFAs in sediments/biofilm/leaf litter samples (64-80% of total PFAS molar concentration); this proportion was lower in invertebrates (28-54%) and in fish (15-26%). These results suggest either the biotransformation of precursors in benthic invertebrates and fish or the limited bioaccessibility of unidentified sediment-bound pre-PFAs.

TH109 PFAS and their precursors in the Environment. First indications from a large scale environmental monitoring study M. Matthoff, Fraunhofer IME Environmental and Food Analysis; M.W. Bücking, Fraunhofer IME Institute for Molecular Biology and Applied Ecology / Environmental and Food Analysis; H. Ruedel, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Department of Environmental Monitoring; J. Koschorreck, Umweltbundesamt Per- and Polyfluorinated Substances (PFAS) have been an ongoing challenge for the environmental sciences for decades. However, the substance versatility, in terms of chemical classes and physico-chemical characteristics yet hinders a full overview of the spectrum. Due to the differential mobility and degradation pathways, the environmental distribution of individual species is complex and requires massive analytical effort. This obscure situation is even stretched by new molecules from international markets, that already travel around the world in order to substitute or as ingredients of rainbow trout applications. Our study set out to apply two large scale multi methods capturing short (e.g. C2 to C6 PFAs), medium and long chain PFAS (e.g. C6 to C14 PFAS and PFPA), and also precursors (e.g. PAPS, dIPAPts, FTS, NaDONA) and novel molecules (e.g. F-35B constituents) on samples of the German Environmental Specimen Bank. Samples include rain samples, suspended particulate matter samples, fish liver, mussels, tree leaves and needles, deer liver, earthworm and herring gull eggs. Here, we present first detections of the F-35B constituents in bream liver samples afar from production sites, and provide indications on distribution patterns.

TH110 A physiologically based toxicokinetic (PBTK) model describing the bioaccumulation of two perfluorinated substances in rainbow trout (Oncorhynchus mykiss) A. Vidal, Istrea Lyon; R. Beaudouin, INERIS / Models for Ecotoxicology and Toxicology METO; E. Vulliet, CNRS / TRACES Team; E. Rochard, Istrea Bordeaux / UR EABX; J. Garric, Istrea Lyon / UR RIVERLAB Laboratoire Ecotoxicologie; J. Maubert, Istrea Lyon / UR RIVERLAB Per- and poly-fluorinated substances (PFASs) are ubiquitous in the environment, specifically in aquatic systems. While several PFASs are acknowledged to be bioaccumulated by vertebrate species, including fish, their absorption, distribution, metabolism and elimination (ADME) remain incompletely understood yet. The aim of this study is to develop a physiologically based toxicokinetic (PBTK) model to predict the bioaccumulation of two selected PFASs in rainbow trout (Oncorhynchus mykiss) exposed through the diet to two selected PFASs, namely perfluorooctane sulfonic acid - PFOS - and perfluorohexane sulfonic acid – PFHxS. Here PFOS is considered as a model compound, as it remains the dominant PFAS in environmental matrices. PFHxS represents an industrial alternative to PFOS, since its addition to Annex B of the Stockholm convention in 2009. Two dietary exposure experiments were performed on adult rainbow trouts (O. mykiss) at two water temperatures (7°C and 11°C). Fish were fed for 96 hpf with contaminated diets spiked with PFOS and PFHxS during several weeks. Then, fish were allowed to depurate, in the same tanks, where non contaminated food was supplied daily. During both phases, 5 randomly selected fish were periodically sacrificed for the analysis of the selected PFASs in muscle, liver and blood. Compound-specific tissue distribution, uptake and elimination rate constants in blood were obtained by a simultaneous adjustment to experimental data. Half-lives were estimated for both compounds, in blood, at both conditions. Globally, fish acclimated to the warmer temperature showed faster absorption and elimination rates of PFOS and PFHxS, and their distribution differed between organs, suggesting that temperature represents an important factor in the toxicokinetic profile of PFASs.

TH112 Toxicokinetics of perfluorinated alkyl acids in zebrafish embryo C. Vego, Karolinska Institutet; G. Johanson, Karolinska Institutet / Institute of Environmental Medicine; M. Nåslund, S. Wulf, Karolinska Institutet / Institute of Environmental Medicine IMM; M. Sjödin, M. Hellström, J. Lindberg, E. Vincent, Swedish Toxicology Science Research Center. Perfluorinated alkyl acids (PFAs) are widely distributed and have been detected e. g. in humans, wildlife and numerous other environmental matrices. These surfactants are highly bioaccumulative as well as persistent and have been associated with several health effects including hepatotoxicity, immunotoxicity and developmental toxicity. The chemical structure of PFAs mainly differ in two ways: the length of the hydrophobic alkyl chain and the hydrophilic end groups. Little or nothing is known how the structure affects the toxicokinetics (TK) (uptake, distribution, biotransformation, elimination) and, consequently, the toxic effects in different organisms. We therefore studied the TK of four PFAs; perfluorooctanesulfonic acid (PFOS), perfluorooctanoic acid (PFOA), perfluorohexane sulfonate (PFHxS) and perfluorobutanoic acid (PFBHA) in the grunion (Sillago sandvicensis), zebrafish (Danio rerio) and rainbow trout (Oncorhynchus mykiss).<ref>TH108</ref> As alternative test model for toxicity testing. We exposed ZFEs at three concentrations of each PFAS up to 120 hours post fertilization (hpf). The test concentrations were selected from pilot studies at which the highest would cause developmental effects in less than 20% of the embryos. Exposure medium and ZFEs were sampled separately at nine time points. Water samples from chemical controls without ZFEs present were taken in parallel. Mass-labelled internal standards specific for each PFAS were added prior to further sample treatment and analysis by LC/MS/MS. The exposure concentrations in the TK experiments differed by four orders of magnitude (PFOS< PFHxS< PFOA< PFBHA). Chemical control concentrations remained constant until 120 hpf for all PFAs tested, ruling out unspecific loss due to adsorption to glass. The time courses of the internal concentrations in ZFE indicated biphasic uptake kinetics with slow uptake before hatching to a faster uptake after hatching. Apparent steady-state concentrations were reached at 96 hpf for PFOS, PFHxS and PFHxS, while PFBHA did not reach steady-state within 120 hpf. Moreover, PFOS and PFHxS (sulfonic acid end group) showed a higher bioconcentration than PFOA and PFBHA (carboxylic acid end group). In conclusion, these data indicate that the functional group of PFAs, in addition to the alkyl chain length, may have an important influence on the toxicokinetic processes.

TH113 Role of bioaccumulation in the derivation of environmental risk limits for two perfluorinated substances, PFOA and HFPO-DA. M. Veugelen, RIVM Institute for Safety of Substances and Products / Centre for Safety of Substances and Products; E. Smit, RIVM / Centre for Safety of Substances and Products; P. Wassenaar, National Institute for Public Health and the Environment (RIVM) Environmental risk limits (ERLs) were derived in the Netherlands for the substances perfluorooctanoic acid (PFOA) and hexafluoropropylene oxide dimer acid (HFPO-DA; also referred to as GenX, FRD-902 or PFPrOPrA). These ERLs serve as advisory values according to the guidance under the Water Framework Directive to set environmental quality standards (EQS) in Dutch policy. For these two PFAS substances, the assessment of the bioaccumulation potential is a key issue in the derivation of the ERLs. The most critical receptors are humans and wildlife, which are not only exposed directly via drinking water, but also obtain a significant part of the total exposure indirectly through their diets. For this purpose, bioaccumulation through the aquatic (and terrestrial) food chains has to be evaluated. This information is amply available for PFOA, but is very scarce for HFPO-DA. For PFOA, a typical bioaccumulation behaviour has been observed.

Absorption, metabolism, distribution and elimination (ADME) are concerned as well. For accurate predictions of organic contaminants bioaccumulation it is therefore important to take into account temperature variations. Also, to our knowledge, no study on the effect of the temperature on the ADME of PFASs in fish has been carried out yet. The aim of this work is to determine to which extent temperature affects absorption and elimination rates, and distribution within the fish of two perfluorinated acid compounds, namely perfluorooctane sulfonate (PFOS) and perfluorohexane sulfonate (PFHxS). Here PFOS is considered as a model compound, as it remains the dominant PFAS in environmental matrices. PFHxS represents an industrial alternative to PFOA, since its addition to Annex B of the Stockholm convention in 2009. Two dietary exposure experiments were performed on adult rainbow trouts (O. mykiss) at two water temperatures (7°C and 11°C). Fish were fed with contaminated diets spiked with PFOS and PFHxS during several weeks. Then, fish were allowed to depurate, in the same tanks, where non contaminated food was supplied daily. During both phases, 5 randomly selected fish were periodically sacrificed for the analysis of the selected PFASs in muscle, liver and blood. Compound-specific tissue distribution, uptake and elimination rate constants in blood were obtained by a simultaneous adjustment to experimental data. Half-lives were estimated for both compounds, in blood, at both conditions. Globally, fish acclimated to the warmer temperature showed faster absorption and elimination rates of PFOS and PFHxS, and their distribution differed between organs, suggesting that temperature represents an important factor in the toxicokinetic profile of PFASs.
The bioaccumulation factors in the aquatic environment appeared to be dependent on the exposure concentration. For both the terrestrial and aquatic food chain, the specific protein-binding behaviour of PFOA requires different methods for normalisation of the concentration values, than those normally applied to hydrophobic substances, i.e. based on lipid and organic carbon. Not only exposure via food, but also the human toxicological threshold value of PFOA itself is dependent on exposure concentration. Higher saturation limits are needed because of the difference in toxicokinetic half-live between human and laboratory animals, like rats and mice. For HFPO-DA kinetic data are only limited, which hampers the derivation of a human-toxicological threshold. It is further investigated based on the available data for both substances whether these findings for PFOA can be extrapolated to HFPO-DA, taking into account the structural differences between both compounds. Additional experimental bioaccumulation data for HFPO-DA is probably needed to complete the ERL derivation.

TH114 Perfluoroether carboxylic acids - are these substances appropriate PFOA-alternatives regarding their environmental concerns? S. Stüben, O.Engler, German Environment Agency - UBA / Chemicals; W. Drost, Federal Environment Agency (UBA) / Chemicals; L. Vierke, German Environment Agency / Chemicals Perfluorooctanoic acid (PFOA) is a persistent, bioaccumulative and toxic substance. To minimize the exposure of humans and environment a restriction according to REACH will come into force in the EU in 2020. For a global restriction, the UVa Limits of the Stockholm Convention. As a result of the regulatory activities as well as voluntary measures, PFOA has been replaced with other fluorinated as well as non-fluorinated alternatives. The use of PFOA as processing aid in fluoropolymer production has been mainly substituted with perfluoroether carboxylic acids (PFECAs). PFECAs are structurally similar to perfluorooakyl carboxylic acids such as perfluorooctanoic acid (PFOA), but they are not bioavailable. Due to this structural similarity it could be expected that PFECAs are equally hazardous to the environment. Thus, the German Environment Agency has assessed the environmental hazards in the context of substance evaluations under REACH for certain PFECAs such as ADONA (ammonium 2,2,3,3-trifluoro-1-(1,2,3,3-hexafluoro-3-trihydroxypropoxy), propionate) and GenX (ammonium 2,3,3,3-tetrafluoro-2-(heptafluoropropoxy) propaneate). The poster will present a summary of the substance evaluations. PFECAs are expected to be very persistent under environmental conditions. The substances have a low bioaccumulation potential in aqueous organisms. However, just as PFOA, PFECAs may not fit into the common accumulation pattern. Furthermore, the substances are probably excreted in the aquatic environment and can reach the ground water and consequently drinking water resources. PFECAs have already been detected in surface water, groundwater and drinking water around fluoropolymer production plants [1-4]. In conclusion, further data are necessary, but the available information on PFECAs already demonstrates that these substances are hazardous for the environment and further risk management measures are needed. [1] Gebbink WA, van Asseldonk L, van Leeuwen ST. 2017. Environ. Sci. Technol. Lett. 4:157-11065 [2] Sun M, Arevalo E, Strynar M, Lindstrom A, Richmond M, Kearns B, Pickert A, Smith C, Knappe DRU. 2016. Environ. Sci. Technol. Lett. 3: 415-419 [3] Schreiber J. 2014. Untersuchung des Transportverhaltens von ADONA in Boden und Grundwasser anhand von Feld- und Laborstudien. Diplom thesis [4] Heydebreck F. T. Jiang J, Xie Z, Ebinghaus R. 2015. Environ. Sci. Technol. 49: 8366-8395 49: 14742-14743

TH115 Fluoropolymers: Polymeric PFAS That Satisfy Global Polymer of Low Concern Criteria B. Henry, W.L. Gore & Associates, Inc. Fluoropolymers, such as polytetrafluoroethylene (PTFE), constitute a distinct class within the polymeric category of the PFAS group. Fluoropolymers are resistant to chemical, hydrolytic, oxidative, photodegradation, and biological degradation. They are thermally stable within their intended processing temperatures (e.g., 260°C for PTFE). Fluoropolymers have negligible residual monomer, low molecular weight oligomers, and negligible intrinsic toxicity. Fluoropolymers with high molecular weights weigh over 100,000 Da, are practically insoluble in water and are not mobile or subject to long-range transport in the environment. Their very high molecular weight prevents fluoropolymers from crossing the cell membrane and thus they are not bioavailable or bioaccumulative. The nontoxic nature of PTFE is supported by numerous Good Laboratory Practice (GLP) studies including acute and subchronic systemic toxicology, irritation on implantation, cytotoxicity, in vivo and in vitro genotoxicity, hemolysis, complement activation, and thrombogenicity. Clinical studies of patients receiving permanently implanted PTFE-containing medical devices demonstrate no chronic toxicity or carcinogenicity, reproductive, developmental or endocrine toxicity. Fluoropolymer medical devices have been implanted in over 40 million patients for over 40 years. This poster includes fluoropolymer biocompatibility/toxicology, human clinical, and physical-chemical-thermal-biological data to show that fluoropolymers satisfy globally recognized assessment criteria to be considered as “Polymers of Low Concern” and to be recognized as being a low hazard class of PFAS. Fluoropolymers, therefore, are distinctly different from the other polymeric and non-polymeric classes of PFAS and should be separated from all other classes of PFAS for hazard assessment or regulatory actions. Grouping all classes of polymeric and non-polymeric PFAS together for restriction or regulation is not scientifically appropriate. Fluoropolymers, as polymers of low concern, are uniquely benign PFAS.

TH116 Fluoropolymers Are Unique, Low Hazard PFAS Needing Different Analytical and Regulatory Approaches Than Monomeric Fluorinated Substances of High Health and Environmental Hazard B. Henry, T. Kennedy, W.L. Gore & Associates, Inc.; H. Fiedler, Otrebro University, Otrebro, Sweden Fluoropolymers, such as polytetrafluoroethylene (PTFE), differ from other monomeric and polymeric per- or polyfluoralkyl substance (PFAS) classes, such as perfluorooalkyl acids, or polymeric precursors that degrade to them. Fluoropolymers do not demonstrate the same toxicity or physical/chemical/thermal properties as other PFAS. Fluoropolymers, such as PTFE do not meet the criteria of PBT (Persistent/ Bioaccumulative/ Toxic) or vPvB (very Persistent/ very Bioaccumulative) chemical substances, nor do they meet the Persistent, Mobile and Toxic (PM or PMT) substances criteria proposed by the German Environmental Agency, Umwelt Bundesamt (UBA, 2017). As high molecular weight fluoropolymers (e.g., PTFE) are benignly persistent (i.e., not mobile, bioaccumulative or toxic), all “highly fluorinated” substances do not pose equivalent health or environmental hazards and thus should not be regulated as a single class of chemicals. Chemical analytical techniques useful for differentiating one fluorinated substance from another are readily available, reliable and reproducible and should be employed to identify and quantify those highly hazardous monomeric per- and poly-fluoralkyl substances (PFAS) individually, rather than techniques aggregating all fluorine containing substances into one group (e.g. total organic halogen, and total organic fluorine). Therefore, high molecular weight fluoropolymers, as a uniquely benign class of polymeric PFAS, require analytical and regulatory approaches differentiating them from fluorine-containing substances that present high health and environmental hazards.

Advances in Soil Ecotoxicology and Risk Assessment of Terrestrial Ecosystems (P)

TH117 Challenges and Open Questions in Earthworm field testing T. Vollmer, Eurofins Agroscience Services EcoChem GmbH / Field Ecotoxicology; O. Klein, Eurofins Agroscience Services EcoTox GmbH / EcoTox Field; S. Kneube, EAS Ectox GmbH / Ectox Field In the risk assessment of plant protection products for in-soil organisms, the earthworm field test following ISO 11268-3 (ISO 2014) is used as the highest tier option. The test protocol is currently under revision and transition to an OECD document under the auspices of UBA (Germany), mainly focusing on improving/testing statistical aspects of the test (e.g. effect of replication) and exploring the options to run the test in a dose-response design. In the light of the recently published EFSA opinion on in-soil risk assessment (EFSA, 2017), and with roughly 20 years of experience with the field test under the ISO guideline, other aspects of the test also might require revision, namely: Description of field site requirements (size and position of initial earthworm population), e.g. minimum requirements, potentially derived from typical MDD values for a given endpoint, Land-use of the field site (arable fields vs. permanent grassland): is there a preferred option, and/or does this depend on characteristics of the substance under test? Site management/maintenance in general (soil cultivation, crop rotation vs. minimum disturbance) and more specifically in the case of testing a substance with herbicidal action (impact on vegetation coverage in test-item treated plots vs. plots of positive and negative control). Plot size and distance between neighbouring plots, plot allocation patterns, and plot separation, especially with a view on potential migration of earthworms between plots and on external re-colonization. Testing of persistent substances (e.g. how to establish a plateau concentration in soil)? Toxic reference (positive control): reduced replication for the toxic reference? Alternatives to the standard reference item Carbendazim? Examples and suggestions will be given and discussed in this contribution and areas for further research will be identified. EFSA [European Food Safety Authority], 2017: Scientific Opinion addressing the state of the science on risk assessment of plant protection products for in-soil organisms. EFSA Panel on Plant Protection and their Residues (PPR). EFSA-Q-2011-00978, Parma, Italy, ISO, 2014: ISO Guideline 11268-3: Soil quality – effects of pollutants on earthworms. Guidance on the determination of effects in field situations.

TH118 Regional Differences of the Environmental Risk Assessment of Pesticides in Soil with a special Focus on the European Union I. Kamoun, National School of Engineering of Sfax, Tunisia / Laboratory of Water, Energy and Environment; J. Sousa, University of Coimbra / Department of Life Sciences; J. Roembke, ECT Oekotoxikologie GmbH
In the European Union (EU) the environmental risk of chemicals is regulated in various ways. Probably the most complex approach in place is the one for pesticides, mainly because these chemicals differ from other chemical groups by three reasons: (1) They are intended to harm organisms, i.e. those which are impacting agriculture. However, many of the pest species affected by pesticides belong to the same taxonomic groups being responsible for many soil functions and services. (2) They are also directly distributed in the environment, usually by spray drift, but also in various other ways such as a coating on seed material. (3) Since their effects are supposed to act only against the pests, when being applied regularly the amount of pesticides ending up in the environment is high. Due to the long experience with this very detailed approach the results of the EU ERA for pesticides are often taken over by other countries (e.g. in Africa). But is this procedure reliable? This contribution focuses on the soil compartment and tries to tackle the following questions: (1) Can (and if yes: how) regional differences (e.g. regarding ecological or agricultural factors) influence the performance or the outcome of pesticide ERA? (2) How do ecological and agricultural differences influence the pesticide ERA within the European Union? Our findings show that regional differences in abiotic, biotic and anthropogenic factors can affect the fate of pesticides in soil as well their effects on soil organisms, meaning that these differences should be considered in pesticide ERA. Proposals will be made how to improve the ERA process but keep it at the same time practical (e.g. by using a tiered approach). These ideas will include the selection of representative reference soils and test conditions for Mediterranean regions. In addition, we will discuss whether the range of standard test species used so far is sufficient. In this context comparisons such used for regulation. This data is also statistically evaluated and helpful. Based on the answers to these questions it will be discussed whether (and if yes, how) the ERA of pesticides has to be modified for Mediterranean regions (both inside and outside of the European Union).

TH119
Adaptation of the earthworm field test method: conceptual overview and first results
J. Roembke, ECT Oekotoxikologie GmbH; B. Danieli, RWTH Aachen University / Institute for Environmental Research Institute; B. Förster, S. Jaensch, ECT Oekotoxikologie GmbH; P. Kotsuik, Umweltbundesamt / Federal Agency of Environment / Risk assessment for plant protection products R. Oettermanns, RWTH Aachen University / Institute for Environmental Research; S. Pieper, German Federal Environment Agency (UBA) / Plant Protection Products M. Ross-Nickoll, RWTH Aachen University, Institute for Environmental Research / Institute for Environmental Research; A. Schieffczyk, ECT Oekotoxikologie GmbH; B. Scholz Starke, RWTH Aachen University / Institute for Environmental Research

In 2016, the German Federal Environment Agency (UBA) launched a project entitled “Necessary adaptations of the standard Earthworm Field Test” to improve the scientific basis when transferring the ISO 11268-3 (1999) guideline to a new OECD standard. As a first step, a literature search was performed to compile available data from earthworm field tests, both from the open literature and from anonymized studies used for regulation. This data is also statistically evaluated and helpful. Based on the answers to these questions it will be discussed whether (and if yes, how) the ERA of pesticides has to be modified for Mediterranean regions (both inside and outside of the European Union).

TH120
Soil ecotoxicology research and ecological risk assessment in southern African mining landscapes
M. Makgoba, North West University / Unit for Environmental Sciences and Management; H. Ejisakkers, WUR/NWU

Although there has been a remarkable development in the field of soil ecotoxicology and risk assessment (RA) models, it is debatable if these RA models are representative enough in order to utilise them on larger spatial scales, attuned to specific landscapes and ecosystems worldwide. An example of this is southern Africa where some soil ecotoxicological research has been done. To address this we will present the results of: An inventory of research on the ecotoxicity of metals in southern Africa which some soil ecotoxicological research has been done. To address this we will present the results of: An inventory of research on the ecotoxicity of metals in southern Africa as an example, focussing on southern African soils, soil life and living conditions. The outcomes of an analysis of the geographical surroundings of gold and platinum mine waste deposits in South Africa The field study of open coal mining in and near a national park in Swaziland It is concluded that there is a limited body of information on southern African soil life, and most of these were laboratory based studies done by a small group of researchers. Future research with regards to incorporating the information available into a soil ecosystem assessment procedure is needed and recommended. It is recommended that a starting point to address this might be the development of site-specific guidelines for Ecological Risk Assessment (ERAs) taking into account landscapes, vegetation and faunal characteristics. From our studies in the surroundings of platinum and gold mine waste, we conclude that these wastes still contain considerable amounts of other chemical elements. The extraction methods, moreover, result in very alkaline or acidic conditions. Further the mine waste is very poorly drained and therefore will provide the conditions to leach out these wastes, given the prevailing wind conditions in these areas, will be dispersed over a wide area causing risks for organisms in natural and built areas surrounding these deposit areas. The coal mine study illustrates that mining in or around natural protected area cause risks due to the irradiating impacts of wind and surface and ground water dispersal form the mined area. Therefore ERA should start to assess the impacts on the natural ecosystems present in the area, and compare these with the outcomes of a Potentially Affected Fraction of species PAF analysis. Key words: soil ecotoxicology, ecological risk assessments, mining, southern Africa

TH121
Establishment of tiered risk assessment approach of pesticides for soil organisms in China
J. Jiang, Nanjing Institute of Environmental Sciences, MEP; J. Zhou, Nanjing Institute of Environmental Sciences, MEP

The aim of the risk assessment for soil organisms is to prevent negative acute or long-term effects on soil functions and functionally most important species. The risk assessment approach established in China considers ecologically relevant groups of soil organisms, e.g. earthworms and soil microorganisms, which are involved in a range of soil functions providing essential ecosystem services, e.g. organic matter breakdown and mineralization, water regulation in soil. The tiered approach is a valuable tool to quickly identify those pesticides which do not pose acute or chronic risk (in a certain area of ecotoxicology) on soil organisms – even under worst case assumptions, and to identify those that need more attention and further evaluations. All risk assessments presented are based on Risk Quotients (RQ), calculated by dividing the Predicted Environmental Concentration (PEC) by the Predicted no Effect Concentration (PNEC). This calculation takes into account, that beside the toxicity of a pesticide, the amount of this pesticide in the environment plays a major role when assessing a risk. If RQ<1, the risk is unacceptable and higher tier risk assessment should be conducted. Exposure and PEC data are only used for tiered risk assessment approach. Tier 1 exposure analysis employs simple model (PECsoil_SFO_China from NIES) to predict exposure to soil organisms. A higher tier exposure analysis can be applied by refining environmental exposure parameters or using semi-field trial test. Currently, the models PRAESS and China-PPEARL, which developed by NIES and ICAMA in China, are applicable to predict the exposure concentration at specific depth of soil layer and at specific scenarios in China. Proposed test systems for effect assessment include acute toxicity test or reproduction of earthworm, reproduction test, nitrogen transformation test, litterbag test and earthworm field test. The PNEC can be calculated using the endpoint obtained from ecotoxicological studies and corresponding uncertainty factors (UF). Tier 1 risk assessment mainly focuses on the exposure of acute or chronic pesticicde to earthworm and soil microorganisms, which are involved assessment and N - transformation assessment. High tier risk assessment mainly focusses on the litterbag test assessment and earthworm field assessment. We have used this tiered risk assessment approach to assess the risk of more than 40 common used pesticides in China. Keywords: tiered, risk assessment, pesticide, soil organism

TH122
Ecological recovery and terrestrial Non Target Arthropods: abundance, functional roles and networks
M. Hagen-Kissling, Eurofins-Mitot; S. Aldershof, Bioresearch and Evaluation; F.M. Thibaut, Eurofins-Mitot

Terrestrial non-target arthropods exhibit a vast array of life history strategies and migration tactics. However, their home range is rather small, and thus, they are a good model group to investigate the factors which influence ecological recovery in risk assessments. Arthropod communities are not stable, but the numbers of species and individuals per species fluctuate over time and space. Part of the variability may be due to the initial disturbances of the application rate of the pesticides which are intrinsic or due to biological interactions, as individuals are embedded in complex food and interaction networks. However, the recovery of a network depends on the fact that all ecological and functional roles within such a complex entity are still realised by a certain number of species (and their individuals). Usually, only abundance criteria are applied in ecological recovery investigations. For example, if numbers of individuals trapped are similar to a control group on two subsequent occasions, recovery is concluded upon. But if the proportion of one functional role (e.g. predators, parasitoids, pollinators or herbivores) in the focal group is far below the proportion of the same functional group in the control group, recovery is not
completed, and the stability of the network in focus might be imbalanced. On the other side, abundance might be different to the abundance in the control group, because of a phase shift due to the initial disturbance, but the proportional distribution of functional roles still mirrors the control group. Thus, we feel that pure abundance data are not enough to understand ecological recovery, but suggest to use additional knowledge about the involved species and their interaction network, like the functional roles and their proportional distribution within a community. Investigating the ecological recovery of a community using information from field work and experiments together with additional information about the species and their importance for and embeddedness in the ecological network is of high importance for a better understanding of the ecological recovery of communities.

TH123 Comparing effects of fludioxonil on non-target invertebrates using ecotoxicological methods from single-species bioassays to model ecosystems A. Hägerbäumer, R. Raschke, Bielefeld University / Animal Ecology; S. Hoss, Ecossa / Animal Ecology; W. Traunspurger, Bielefeld University / Animal Ecology Lower tier toxicity testing used for risk assessment of plant protection products (PPPs) is conducted with single species. Information from such toxicity tests are, however, limited to direct effects of the respective tested substances. Although the uncertainties regarding the protectiveness of these tests for in-situ communities are known, informations on effects of PPPs on community-level of non-target organisms are scarce. Model ecosystems, i.e. microcosms, are suitable to bridge this gap between single-species tests and field studies since they provide controlled experimental conditions and are able to demonstrate direct and indirect effects of the respective substances. In the present study, single-species toxicity tests and soil-spiked microcosms were used to comparatively investigate the toxicity of the non-systemic fungicide fludioxonil (FDO) on non-target soil organisms. Regarding soil invertebrates, nematodes are among the most abundant metazoon organisms and hence more often used as components in soil communities are suitable for risk assessments via various assay tools ranging from single-species toxicity tests to field studies, potential effect of FDO on these non-target organisms were assessed using standardized toxicity tests with Caenorhabditis elegans (ISO 10872) in spiked soil exposure and in-situ nematode communities, sampled from microcosms with FDO-spiked soils. In the standardized toxicity tests, FDO inhibited the reproduction of C. elegans dose-dependently, with a chronic 96-h EC50 of 363 mg kg⁻¹ (dry weight, dw) and a 96-h No Observed Effect Concentration (NOEC) of -1 (dw), which is comparable to the 28-d NOEC of Chironomus riparius in sediment (40 mg kg⁻¹ dw) and within the range of the 56-d NOEC of Eisenia fetida (20 mg kg⁻¹ dw) and 28-d NOEC of Folsomia candida (125 mg kg⁻¹ dw). In the spiked microcosms, distinct effects on in-situ nematode communities could be measured, with significantly lowered abundances in spiked soils, (40% and 50% reduction of nematode abundance in soils spiked with 300 and 600 mg FDO kg⁻¹ dw). Overall, this study provides new insights into the impact of the non-systemic fungicide fludioxonil on non-target soil organisms and demonstrated the general suitability of standardized toxicity testing on C. elegans in protecting in-situ communities.

TH124 To what extent do soil micro-arthropods facilitate OM breakdown in an arable field soil? - Implications on specific protection goal setting for soil risk assessment of plant protection products G. Lewis, JSC International Ltd; S. Brauker, BASF France S.A.S.; C. Mayer, BASF SE / Ecotoxicology

The concept of source-sink dynamics as a potentially important component of metapopulation dynamics was introduced in the 1980’s culminating in the paper by Pulliam (1988). Since that time, a considerable body of work has developed to consider its theoretical implications as well as to identify how it may be manifested under field conditions. Most recently, the concept of source-sink dynamics has been considered within the European Food Safety Authority (EFSA) Opinion addressing the state of the science on risk assessment of plant protection products for non-target arthropods (EFSA, 2015). This presentation reviews the available literature that investigates the theoretical implications of source-sink dynamics as well identifying the relevant available evidence from both experimental systems and field observations, primarily in relation to non-target arthropods in an agricultural environment. Consideration of this information clearly shows that metapopulation dynamics are generally more complex than presented by the simple source-sink model as originally proposed and that they are very much species/context dependent. However, this issue does raise important questions for toxicologists and risk assessors when dealing with soil organisms and further investigation is needed in order to better understand the role and influence of differences in landscape structure on the population dynamics of non-target arthropods in the context of the risk from the use of plant protection products. It is therefore important to consider what the evidence base is for source-sink dynamics in the agricultural environment and what this tells us about whether or how it is manifested in relevant populations. A structured approach can then be adopted in terms of identifying suitable surrogate species and generating the necessary information for them and at the landscape scale to allow the development of suitable population models. These models could then be used in an appropriate way within a risk assessment scheme e.g. at a higher tier level addressing specific issues of concern identified at the lower tiers. They may also have the potential to inform risk managers to consider the scale of the environmental footprint of their activities.

TH125 Classification of uncertainty in ecological risk assessment of pesticides A. Hunka, Halkstad University / School of Business, Engineering and Science; M. Meli, ADAMA Agricultural Solutions; S. Pashami, S. Waara, Halkstad University

Uncertainty estimates are inherently built into any prospective risk assessment. Uncertainties need to be correctly recognized, described and presented to provide a basis for decision-making. One important factor to consider is that more data and experimental results often lead to an increased perception of uncertainty. Practical experience indicates that although the amount of information needed in relation to the life-cycles of non-target arthropod species and landscape structure, are also identified. Acknowledgements: This work was initiated and funded by the ECPO non-target arthropod group.

TH126 Classification of uncertainty in ecological risk assessment of pesticides A. Hunka, Halkstad University / School of Business, Engineering and Science; M. Meli, ADAMA Agricultural Solutions; S. Pashami, S. Waara, Halkstad University

Uncertainty estimates are inherently built into any prospective risk assessment. Uncertainties need to be correctly recognized, described and presented to provide a basis for decision-making. One important factor to consider is that more data and experimental results often lead to an increased perception of uncertainty. Practical experience indicates that although the amount of information needed in relation to the life-cycles of non-target arthropod species and landscape structure, are also identified. Acknowledgements: This work was initiated and funded by the ECPO non-target arthropod group.
Derivation of soil threshold concentrations for arsenic: consideration of bioavailability through combination of ecotoxicological and analytical data


The German Federal Soil Protection Act (1988) defines precautionary values for several metals which, if exceeded, indicate that concern for a harmful soil change exists. All precautionary values given in the German Soil Protection Ordinance (1989) are based on total concentrations ("aquaria"). However, a realistic risk assessment of metals should consider their bioavailability in soil. Thus, the aim of this project is to connect bioavailable fractions of arsenic with ecotoxicological effect concentrations, taking into account soil properties (texture, pH, organic matter content etc) and various metal extractions (1M NH4NO3, 0.10M CaCl2, Ca(NO3)2; with ionic strength corresponding to soil solution, DTPA/CaCl2, 0.43M HNO3, plus aqua regia). Arsenic was chosen due to its high relevance as a soil contaminant, its low data availability compared to other metals and is an element of concern included in many soil regulations. Six soils covering a wide range of Central European soil properties were chosen and spiked with sodium arsenate dibasic (Na2HAsO4.7H2O). Chronic toxicity endpoints were tested with microbes, plants and invertebrates, according to ISO standard guidelines, allowing derivation of threshold values via an SSD approach. The results (given as NOEC, EC10 and, preferably, EC50 values, based on the six extraction methods, have been determined. The variation in EC10 values based on nominal concentrations among the soils tested differed typically by a factor of 2 - 5 for the endpoints tested. The extraction strength of the different methods and soils differ at least by an order of magnitude in the order NH4NO3 < CaCl2 < Ca(NO3)2 < DTPA < aqua regia. However, most of the most sensitive, partly together with the Bacteria. Both invertebrate species were always less sensitive (i.e. EC50 values (nominal concentration) > 250 mg as/kg soil) than microbes and plants except in one sandy soil (RefSoil-01A). Currently, chemical and biological results are combined in order to explain the observed variation in toxicity expressed as nominal or total As concentrations in soil. This information will be used to include As bioavailability into the derivation of precautionary values. The representativeness of the different extraction methods regarding bioavailable fractions as well as the properties of the different soils are checked as a part of a more realistic risk assessment of metals in soils.

Activity based in-soil arthropod sampling

S.B. Dehlehan, F.M. Bakker, Eurofins-Mitox

Higher tier (field) assessment of effects on soil microarthropods relies strongly on the accuracy of the sampling methodology. Two main classes of trapping methods exist to date, these are either abundance-based or activity-based. Abundance-based trapping directly involves the collection of soil arthropods followed by heat extraction such as Berlese-Tullgren or McCafayed methods. Activity-based sampling implies installing hypogeae traps and collecting the catch at pre-determined intervals. Soil core sampling provides an instantaneous assessment of the fauna at the exact moment and at the very location of sampling, whereas hypogeae traps provide an assessment of the activity in a wider area and over a longer time span. Clearly, higher tier assessment of abundance activity is then used to determine the nutrient activity. Soil core sampling is an established and recommended method known to extract spingtails, mites and some other small arthropods. Hypogeae trapping is a relatively novel approach (cf Dehlehan et al. SETAC 2016, Sims et al. 2016, Bakker et al. 2017) and seems to have a certain degree of selectivity. For purposes of method development and evaluation we have performed a comparative study in which soil core sampling and hypogeae trapping (mine traps) were performed in the same fields. The study comprised both a hay meadow and an arable field. Soil cores were taken from the top 10 cm of soil, mine traps collected from various depths. With this contribution we will highlight the differences in species spectrum, numbers collected and variability observed with the different methods and discuss the implications for data analysis and interpretation.

The application of the CPCAT approach reduces shortcomings of effect detection for earthworm field studies

B. Daniels, RWTH Aachen University / Institute for Environmental Research; S. Jänsch, ECT Oekotoxikologie GmbH; P. Kotschik, Umweltbundesamt / Federal Environmental Agency / Risk assessment for plant protection products; R. Ottermanns, RWTH Aachen University / Institute for Environmental Research; S. Pieper, German Federal Environment Agency (UBA) / Plant Protection Products; M. Ross-Nickoll, B. Scholz-Starke, RWTH Aachen University, Institute for Environmental Research / Institute for Environmental Research Field studies to determine effects of pollutants on earthworm community are generally conducted according to standardized ISO-guidelines (ISO 11268-3). However, statistical test procedures suggested in the guidelines are frequently criticized, mainly for two reasons: test data characteristics do not fulfill test requirements (normal distribution and variance-homogeneity) and the resulting toxicity metrics of multiple testing procedures (NOEC / LOEC) fail to adequately detect the actual level of effects. Lehmann et al. (2016) presented a new approach to overcome these shortcomings by introducing the CPCAT procedure. We applied this statistical method to detect effects in a set of 16 earthworm field studies and provide a comparative analysis with regard to results of well-established multiple testing approaches. This allows us to compare the performance of CPCAT with an extensive meta-analysis of field study data. Raw data of biomass and abundance on sample level (0.25 square metres) were extracted from original study reports and assessed on sample and plot level. In total, data of 17 different earthworm species, ecological and morphological groups as well as total abundance and biomass for 1-3 treatments and 3 sampling dates after application within test duration of one year were analysed. This led to a total of 4215 comparisons for the detection of differences between control and treatments. We demonstrate that the distribution of both endpoints abundance and biomass can be described by a Poisson model, which is a requirement for the application of CPCAT (variance homogeneity -often not fulfilled in toxicity tests- is not a prerequisite of CPCAT). The number of endpoints showing a significant difference between control and treatment was compared to the outcome of parametric test procedures (pairwise t-test, Dunnett and Williams t-test for multiple testing). The study reveals that the application of standard multiple testing procedures leads to a disguising of possible effects due to relatively high differences to be achieved between control and treatments. This consequently results in uncertainties regarding the actual level of effects at the NOEC. The CPCAT approach offers a more powerful and statistically proper alternative to the determination of field studies because data distribution and variance are adequately considered and smaller differences between control and treatments can be detected.

Relationship between soil microbial biomass methods used in environmental fate laboratory studies

P. Massey, Smithers Visicient; P. Pearson-Davies, B. Earnshaw, Smithers Visicient; S. Swales, Smithers Visicient ESG Ltd

The determination of microbial biomass activity is a pre-requisite for OECD laboratory studies that are designed to investigate the environmental fate of chemicals in soils. By determining soil microbial biomass prior to, during and after a study, the viability and suitability of the experimental soil can be demonstrated. Choosing the appropriate methodology for soil microbial biomass is critical for conducting successful environmental fate studies. One common method, referenced in the OECD guidelines, for determining soil microbial biomass is the fumigation extraction method. This method determines the carbon content of the soil biomass, via fumigation, using ethanol-free chloroform. During fumigation, cells are lysed by the chloroform, which results in a flush of organic carbon into the soil environment. This organic carbon is then extracted and quantified. Another suitable way of estimating soil microbial biomass is by substrate induced respiration. This method uses a suitable labile substrate to promote a respiratory response. The carbon dioxide evolved or the oxygen consumed as a result of this respiratory response is then measured. With both methods, the biomass size can then be determined by relating respiration and fumigation extraction data. In spite of these multiple recognised ways of determining soil microbial biomass, it is important to recognise that they reflect different aspects of the soil microbial community. One fundamental difference between these methods is that they can potentially distinguish between active and non-active components of the biomass. As noted in OECD guidelines, substrate induced respiration can be used to estimate the active aerobic biomass, whereas in OECD 14240:2:1997, fumigation can extract carbon from both active and non-active biomass components. With such differences between methods, it is important to consider which method is more appropriate for determining soil suitability for environmental fate laboratory studies. In work currently being undertaken by Smithers Visicient to investigate the relationship between the soil microbial methodologies commonly used for laboratory soil studies. The aim of this work is to better understand how the choice of soil biological methods relates to soil suitability, which will ultimately facilitate and refine our choice criteria when choosing soils for environmental fate studies.
The population dynamics experiment is a one-year study assessing the dynamics of *Folsomia candida* in artificial OECD soil at constant 20°C. The study started with 25 individuals of different age classes in 100 g OECD soil. Since then the population increase was measured on at least a monthly basis with five replicates per testing day. The food regime is adapted to the increasing population density to make sure that the maximum population level is achieved during the study. We will show a fast growth at the beginning of the experiment and expect to reach an oscillatory pattern in its maximum by the end of the study. In a second experiment the vertical dispersal of *F. candida* in relation to food location is investigated. Transparent PVC columns were filled with on average 350 g of OECD soil up to a level of 20 cm soil column height and 86 *F. candida* of different age classes. Each column was closed with Parafilm on top and a gauge on the bottom with a small water reservoir beneath it to avoid desiccation. The initial humidity was set to 50 percent of the maximum water holding capacity of the soil. The columns were separated into six compartments at different heights: 1, 2.5, 5, 10, 15 and 20 cm. Sepht in the middle (4th) and bottom (6th) compartment allowed for watering and feeding. We varied the location of feeding by four different regimes while all other parameters were kept constant. The columns were provided either with food at the top in the middle, in the ground or at all three compartments. Our hypothesis is that food is a main trigger for the vertical distribution of *F. candida* in soil. Two of three large examination dates have been processed so far. The data confirms our hypothesis and the results of the study will present new data for the otherwise well investigated Collembola species *F. candida*.

**TH132**

**Why zinc doesn't matter: habitat quality drives invertebrate response to zinc, not concentration**  
S. Siciliano, University of Saskatchewan / Department of Soil Science; K. Jegede, H. Fujana, University of Saskatchewan Toxicology Centre  
The responses of organisms in soil ecotoxicity tests are often determined by the bioavailable concentrations of contaminants they are exposed to. However, the direct effect of habitat quality on the performance or response of organisms in different contaminated soils is often neglected. Habitat quality is a measure of extent to which habitat promotes individual and population fitness. This study assessed the effect of habitat quality on mite, *Oppia nitens* exposed to different contaminated soils which was corrected for bioavailable metals. Forty-seven (47) soils were ranked into habitat quality by summing up the scores of enchytraeid and collembola survival and reproduction with the plant biomass in each of the soils. From the 47 soils, 18 soils were divided into three habitat quality groups based on high, medium and low habitat quality. The 18 soils were dosed with low to high concentrations of zinc and mites exposed to the soils for 28 days. Mite survival, reproduction, food at the top, in the middle, in the ground or at all three compartments. Our hypothesis is that food is a main trigger for the vertical distribution of *F. candida* in soil. Two of three large examination dates have been processed so far. The data confirms our hypothesis and the results of the study will present new data for the otherwise well investigated Collembola species *F. candida*.

**TH134**

**Effects of atmospheric hydrogen chloride and ammonia on Paronychiurus kimi (Collembola : Onychiuridae)**  
J. Weg, J. Son, Korea University / Division of Environmental Science and Ecological Engineering; Y. Kim, Y. Lee, J. Hong, M. Lee, Korea University; K. Cho, Korea University / Division of Environmental Science and Ecological Engineering  
As the use and distribution of various chemicals increases, there is a possibility of chemical accumulation in Korea, the incidence of chemical accidents is also increasing. Damage caused by chemical accidents is not only widespread but also has a long-term impact, making it difficult to predict damage and respond appropriately however, there are very few studies on chemicals that can cause accidents. Especially for chemicals exposed to gaseous state, little is known about the impact of soil biocenoses such as Collembola and earthworm. The experiment was carried out in PS container filled with soil according to modified OECD 232 guidelines. Investigating the effects of gaseous hydrogen chloride and ammonia on *Paronychiurus kimi* (Collembola), the test vessels with *P. kimi* were exposed to two different concentrations of toxic substances in the enclosed chamber for 20 minutes. After exposure, the test vessels with *P. kimi* were transferred to an incubator (20°C, continuous darkness) in a closed state, and the mortality and reproduction rate of *P. kimi* were observed after 1 hour, 2 weeks, and 4 weeks. There were no deaths after 1 hour, but the mortality rate was increased over time from 2 weeks. After, 4 weeks, the number of juveniles produced by adults *P. kimi* were decreased as concentration-dependent manner. These results show that the long-term effects of gaseous phase chemicals can occur at concentrations that are not acutely affected.

**TH135**

**Toxicity assessment of methyl ethyl ketone using earthworm and soil algae**  
R. Cui, Konkuk University / Department of Environmental Sciences; Y. An, Konkuk University / Department of Environmental Health Science  
Methyl ethyl ketone (MEK) is a kind of ketone-based volatile organic compound and widely used as industrial solvent. There is a high possibility of leakage of this compound into soil environment, but few studies for ecotoxicity of MEK were present. This study evaluated the toxicity of MEK using earthworm *Eisenia andreii* and algae *Chlamydomonas reinhardtii* and *Chlorococum infusionum*. *Eisenia andreii* were exposed with 1% of control or MEK soils with closed system. After 7 days exposed, mortality and abnormalities including bloating, fragments, swelling, bleeding, and mucous secretion were measured. For soil algae, *Chlamydomonas reinhardtii* and *Chlorococum infusionum* were exposed with 2.5 g of control or MEK soils in 15 mL glass test tube, and chlorophyll intensity was measured after 6-day exposed. As results, 7d-LOEC and 7d-EC50 of MEK to *Eisenia andreii* were calculated as 1136 mg MEK/kg dry soil and 1910 (1643.00-2221.58) mg MEK/kg dry soil, respectively. For soil algae, *C. infusionum* was more sensitive than *C. reinhardtii* for MEK. 6d-EC50 to *C. reinhardtii* and *C. infusionum* were calculated as 3400.44 (3132.01-3690.94) mg MEK/kg dry soil and 60.97 (51.19-72.62) mg MEK/kg dry soil, respectively. These results can be used for risk assessment of MEK in soil ecosystem. *This work was supported by Korea Environment Industry & Technology Institute (KEIT) through "The Chemical Accident Prevention Technology Development Project”, funded by Korea Ministry of Environment (MOE)(No. 2016001970001).* **<strong>Key word: methyl ethyl ketone, earthworm, soil algae**

**TH136**

**Effects of endocrine disrupt chemicals (EDCs) to soil algae**  
R. Cui, Konkuk University / Department of Environmental Sciences; Y. An, Konkuk University / Department of Environmental Health Science  
There were many data for endocrine disrupt chemicals (EDCs) for aquatic organism, but soil toxicity data of them were very limited. This study evaluated the effects of bisphenol A (BPA), bis(2-ethylhexyl)phthalate (DEHP), and nonylphenol used for test the effects of chemicals. Soil algae *Chlamydomonas reinhardtii* and *Chlorococum infusionum* were exposed at 0.5 g of control or exposed soils in 6-well plate. Algae were extracted for 1 day using algae culture medium after 6-d exposure, and the chlorophyll intensity was measured by fluorescence microplate reader. We observed that the BPA was most toxic following NP and DEHP. The effect of DEHP was insignificant to *Chlamydomonas reinhardtii* and *Chlorococum infusionum*. The results can be used for risk assessment of BPA, DEHP and NP in soils. This study was funded by the Korea Ministry of Environment (MOE) as the Environmental Health Action Program (1485014458) **Key word: bisphenol A, bis(2-ethylhexyl)phthalate, nonylphenol, soil algae**

**TH137**

**Evaluation of reproduction tests of earthworms and enchytraeids exposed to sugar cane vinasse in natura and after pH adjustment**  
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The use of sugarcane vinasse as fertilizer in crops has been widely used in order to provide an adequate destination for this residue however, it has properties that can be prejudicial to the animals present in the soil, as already verified in numerous studies. Therefore, the objective of this work was to treat vinasse with lime (CaO) to adjust pH to 7.0 (neutral), in an attempt to reduce its toxicity for later use in the soil. In this context, the development of ecotoxicological tests presents itself as a tool of great assistance in the analysis of residues released to the soil. Thus, reproduction tests were conducted using animal soil biocenotes to evaluate the effects of vinasse may have on reproductive behavior of these animals. Earthworms of the species *Eisenia andreii* (Annelida) and enchytraeids of the species *Enchytraeus crypticus* (Annelida); both tests were developed according to the protocols proposed in ISO 11268-2 (ISO, 2011) and ISO 16387 (ISO, 2013), respectively. In the reproduction tests with *E. andrei*, soils amended with vinasse in natura in comparison to the animals exposed to the treated vinasse there was an increase in the number of animals, which suggests that the vinasse treatment for pH adjustment was valid for this test. The reproduction test with *E. crypticus* exposed to the same conditions cited above also showed an increase in the number of individuals exposed to treated in biosassays vinasse compared to exposed to vinasse in natura. The results allow to infer that the pH adjustment of the vinasse to a neutral level was effective in reducing the toxicity of the residue for the tests of reproduction both in species used, since the environment favored the reproduction of the animals tested.

**TH138**

**Ecotoxicological Characterization of Nitrogen-Based Energetic Soil Contaminants**  
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Environmental Toxicology

We provide an overview of ecotoxicological effects of nitrogen-based energetic materials (EM) of notable ecological concern, hexahydro-1,3,5-trinitro-1,3,5-triazine (RDX), octahydro-1,3,5,7-tetranitro-1,3,5,7-tetrazocine (HMX), and 2,4,6,8,10,12-hexanitro-2,4,6,8,10,12-hexaazaowurzitane (CL-20), 2,4,6-trinitrotoluene (TNT), 1,3,5-trinitrobenzene (TNB), 2,4-dinitrobenzene (2,4-DNT), 2,4-dinitrotoluene (2,4-DNT), 2-amino-4,6-dinitrotoluene (2-ADNT), 4-amino-2,6-dinitrotoluene (4-ADNT), and nitroglycerin (NG). Ecotoxicological effects of these EMs were determined in definitive studies with Sassafras sandy loam (SSL) soil using soil invertebrate, terrestrial plant, and biological activity endpoints. SSL soil was selected because it has physicochemical characteristics (low organic matter and clay contents) that support very high relative bioavailability of organic chemicals. Data for reproduction (soil invertebrates), growth (plants), and critical soil processes (basal and substrate-induced respiration, litter decomposition) were independently analyzed using appropriate regression models to determine the EM concentration producing 20 percent decrease (EC20) in the measurement endpoint compared with carrier (acetone) control. Toxinological benchmarks developed in studies with soil invertebrate and terrestrial plants were used to derive draft Ecological Soil Screening Levels (Eco-SSLs) for use in screening-level ecological risk assessment of EM-contaminated soils.

Additionally, we developed species sensitivity distributions (SSDs) for select EMs using toxicity data for all three soil ecological receptor groups (invertebrates, plants, and soil processes). These SSDs were then used for derivation of Soil Contaminant Scores (SCSs). Bacterial strains at 200 mgkg\(^{-1}\) of EM were inoculated into copper oxychloride-spiked soils and earthworm tissues were determined. Findings revealed that both transformed and un-transformed \(E.\ coli\) JM109 were used to derive specific SCV protective of plants, soil invertebrates, and critical soil processes.

TH139 Organismal responses of oligochaetes in bacterial inoculum amended copper oxychloride spiked soils

M. Maboeta, North-West University / Unit for Environmental Sciences and Management; O. Oladipo, M. Engelbrecht, North-West University

The excessive release of heavy metals such as copper via anthropogenic sources into the soil environment has raised some global concern. Copper oxychloride, a common agricultural metal-based fungicide applied to fungicidal crops, contains 60% copper. This high copper content may significantly contribute to the soil copper burden and negatively affect the mesofauna.

Metal-tolerant bacteria such as \(Bacillus\) cereus strain have been identified for their bioremediative traits in metal polluted soils. We examined the effect of \(Achromobacter\) sp - \(Bacillus\) cereus consortium on the ecotoxicity of copper oxychloride spiked soils. Bacterial consortium isolated from copper oxychloride spiked soils and the terrestrial bacteria strains used in this study, the bacterial strains used (\(Achromobacter\) sp and \(Bacillus\) cereus) were previously isolated from soil and gemstone mining sites and confirmed to tolerate up to 200 mgkg\(^{-1}\) Cu. Twenty-four hours pure broth cultures of the two bacterial strains were inoculated into fungicide spiked soils. Utilizing standard ISO and OECD protocols, 10 mature \(Enchytraeus\) and \(Enchytraeus\) albida were exposed separately to both inoculated copper oxychloride and uninoculated copper oxychloride spiked soils for 28 days. Avoidance behavior, biomass, reproductive success, metal contents in soils and earthworm tissues were determined. Findings revealed that \(E.\ andrei\) in inoculated substrates (200 mgkg\(^{-1}\)) exhibited significantly higher (p < 0.05) preference, relative growth rate, survival, cocoon and juvenile counts and soil Cu content (comparable to the control) than non-inoculated soils. Similarly, with the \(E.\ albida\) significant higher (p < 0.05) preference and reproductive success was recorded. However, at 1000 mgkg\(^{-1}\) copper oxychloride soils, no distinct effect was observed on both \(E.\ andrei\) and \(E.\ albida\) in bacterial inoculated and non-inoculated substrates. In conclusion, \(Achromobacter\) sp - \(Bacillus\) cereus bacterial consortium demonstrated the ecotoxicity of metal-based fungicide towards \(E.\ andrei\) and \(E.\ albida\). Copper oxychloride spiked soils at 200 mgkg\(^{-1}\). \(Achromobacter\) sp and \(Bacillus\) cereus are therefore recommended for the bioremediation of soil contamination of copper contaminated environments. Keywords: Copper oxychloride fungicide. \(Achromobacter\) sp - \(Bacillus\) cereus consortium. Ecotoxicity. Oligochaetes

TH140 Development of a terrestrial biotic ligand model (TBLM) for predicting acute toxicity of cadmium and zinc to soil collemboles Paronychiurus kimi

J. Son, K. Cho, Korea University / Division of Environmental Science and Ecological Engineering

Complex interactions between metals and soil properties make it difficult to apply a biotic ligand model widely used in aquatic ecotoxicity studies. In this study, a terrestrial biotic ligand model (TBLM) was developed to predict the acute toxic effects of cadmium and zinc on the survival of soil collemboles Paronychiurus kimi in a simplified soil solution system under the assumption that soil pore water is the main route of exposure to metals. After 7 days of exposure, survival and internal metal concentrations in \(P.\ kimi\) were determined. The free metal ion activity for each metal was calculated by the Visual MINTEQ using inputs of soil metal concentrations, cation and anion components of the soil solution, and pH of the soil solution. The toxicity of cadmium and zinc was linked to the fraction of biotic ligand occupied by metal, and the free metal (i.e. \(\text{Cd}^{2+}\) and \(\text{Zn}^{2+}\)). The results showed that the ratio of the biotic ligand occupied by metal can be predicted to the metal toxicity, indicating the applicability of TBLM to explain metal toxicity to \(P.\ kimi\) in a simplified soil solution. Although the approach used in this study may be limited to soil solution, the use of TBLM can be a useful tool for investigating factors affecting bioaccumulation and toxicity of metals.

TH141 Characteristics of metal-tolerant bacterial plasmids from a platinum mine tailings dam

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The presence of mine tailings often promote the development of both heavy metal and antibiotics resistance among microbes with resistant plasmids. Plasmids provide their hosts with a large array of phenotypes such as heavy metals and antibiotics resistance due to gene transfer. This study describes the characteristics of plasmids isolated from various bacteria that displayed an ability to withstand high metal concentrations. Isolated plasmids were individually transformed into \(Escherichia\) coli \(JM109\). The plasmids were evaluated for metal tolerance capabilities using a microdilution approach where the plasmid DNA concentration ranged between 11.75-118.06 ng/µl after extraction. Incompatibility groups were determined by subjecting plasmids to PCR amplification using Inc\(Q\), Inc\(P\) and Inc\(W\) specific primers, where only Inc\(W\) provided positive results. Minimum inhibition concentrations (MICs) were carried out to determine the ability of transformed \(E.\ coli\) JM109 to tolerate metals at varying concentrations. Results indicated that transformed \(E.\ coli\) JM109 developed ability to grow in the presence of several heavy metals. Some strains were resistant to high concentrations (+10 mM) of \(\text{Ni}^{2+}/\text{Pb}^{2+}/\text{Ba}^{2+}\) with metal resistance order of \(\text{Ni}^{2+}/\text{Pb}^{2+}/\text{Ba}^{2+}\). Moreover, protein profiling was used to determine the impact of plasmids on \(E.\ coli\) JM109. Proteins were extracted from both transformed and un-transformed \(E.\ coli\) JM109 and subjected to one-dimensional (1D) and two-dimensional (2D) SDS-PAGE. One dimension SDS-PAGE illustrated general similarity of the profiles except for two banding positions in the 30 to 35 kDa region where bands were present in the transformants that were grown in the Al/Ni alloy containing media. Two-dimensional electrophoresis PAGE and 2D electrophoresis PAGE showed that some of the proteins were up-regulated while others were down-regulated. The largest numbers of proteins were from 15 – 75 kDa. Since the plasmids rendered the \(E.\ coli\) JM109 tolerant to metals, it can be concluded that the change in the protein profiles was due to the effects of the plasmids. Furthermore, that plasmids isolated from various heavy metal-tolerant bacterial species were successfully transformed into \(E.\ coli\) JM109 rendering the \(E.\ coli\) JM109 tolerant to metals. This study shows that plasmids characterized have advanced our understanding that these plasmids could be important reservoirs for resistant genes, and may hold significant biotechnology potential.

TH142 Sensitivity of the waterside species, Yuukkanura szepczykii (Collembola: Neanuridae), to cadmium and copper

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Collembola is the most abundant organism in the soil ecosystem and some species are used as ecotoxicological evaluation species for toxic substances in soil. However, Neanuridae species, which is one of the family of Collembola, are rarely studied as toxicity evaluation species. In this study, the toxicity sensitivity of copper and cadmium of \(Yuukkanura szepczykii\), known as the species in which they live water, and their biomagnification in soil were examined based on the ISO guideline 11267. After 28 days of exposure to tested metals, LC50 for adult survival and EC50 for reproduction were estimated. These toxicity values of \(Y.\ szepczykii\) were also compared to those of other collemboles species (\(F.\ candida\) and \(Paronychiurus\) kimi) reported in literature to investigate their suitability as a new test species in toxicity test. Not only the adult survival but also the juvenile predation of \(Y.\ szepczykii\) was observed in a collection dependent manner after 28 days of exposure duration. Although the response of \(Y.\ szepczykii\) to the tested metals was not highly sensitive to the other collemboles species reported in literature, the study of the response of \(Y.\ szepczykii\) to chemicals in the soil is considered to be very important. Because their special habitat can provide an understanding of ecotoxicity against certain environmental conditions.

TH143 Drivers of copper and zinc availability and phytovailability in agricultural soils receiving long-term organic waste amendments

SETAC Europe 28th Annual Meeting Abstract Book
The potential ecotoxicological effects of this contamination on soil organisms depend on Cu and Zn availability in soils. The availability of Cu and Zn itself depends on their chemical speciation and consequently to the temporal evolution of soil parameters such as pH and organic carbon content and reactivity. These soil parameters are both influenced by the application of OW and the activity of soil organisms on the surrounding soil, i.e. the rhizosphere for plants. However, the detailed influence of OW applications on the rhizosphere effect are poorly documented when taking into account long-term impacts. Accordingly, we aimed at studying the relationship between the availability in soil and the phytoavailability of Cu and Zn in four decadal field trials that received different types of OW for more than ten years. Soils in the four field trials exhibited very different pH and organic carbon content. Copper and Zn availability was determined on 102 soil samples from the four field trials by (i) an equilibrium-based method using cupric ion selective electrode and the winer the hemic aqueous model (WHAM) to quantify Cu2+ and Zn2+ activities in soil solutions (pCu2+ and pZn2+) and (ii) a kinetic method using the diffusive gradient in thin films (DGT) directly on soil samples. We measured key soil parameters in soil solutions to assess the relationship with pCu2+ and pZn2+. Copper and Zn phytoavailability is currently determined using the RHIZOtest which is a standardized biotest that is used to measure the uptake flux of Cu and Zn in plants and the related availability of Cu and Zn in the rhizosphere that is physically separated from roots. The results already achieved showed no clear relationship between Cu2+ and pH or dissolved organic carbon among the four field trials altogether. When studying each trial separately, we observed a pH gradient as a function of the type of fertilizer (mineral or OW) applied in the treatment. Additionally, the use of kinetic measurements will enable to test whether Cu and Zn availability in unplanted soils determined Cu and Zn phytoavailability or whether root-induced chemical changes in the rhizosphere additionally determined it.

The fate and bioavailability of currently used and emerging pesticides in agriculture were studied. 

**TH144** Toxic Effects of Cadmium on Chinese Cabbage, *Foliosima Candida*(collembola) and their Prediction Modes in 18 Soils of China

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In this paper, we adopted 18 Kinds of typical soils in China, and Chinese cabbage, *Foliosima candida*(collembola) were used as the research object. The germination and root elongation of cabbage under different concentration of cadmium in soil were measured. The endpoint of the F. candida was reproduction. The results show that the soil properties significantly affected the dose effect curve of cadmium, soil pH is the main influencing factor; at the same time, we calculated the toxicity threshold and prediction models. This study has a guiding significance for the plant and invertebrates ecological risk prediction and assessment of heavy metal cadmium.

**TH145** Do we plant protection products correctly? Impact of agrochemicals on non-target beetles, *Bembidion lampros*(Coleoptera: Carabidae)

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Intensification of agriculture and the widespread use of pesticides during the last few decades have led to significant reduction of the abundance of non-target arthropods (NTAs), including the ground beetles (Carabidae), which are natural pest enemies in agricultural areas. Due to the growing demand for food, it is not possible at the moment to stop using pesticides. We need, therefore, to make every effort to ensure that they are used in a way that do not jeopardize NTA. In the present study, three commonly used pesticide formulations: Durban, 480 EC, containing the organophosphate parathion, and the pyrethroid esfenvalerate (CYP), and Spekfree 430 SC, containing the fungicide tebuconazole (TEB), were tested for their effects on survival of the ground beetle *Bembidion lampros*. The beetles were collected from agricultural fields either in spring (after overwintering) or autumn (population dominated by newly emerged individuals) and exposed individually to a single pesticide spray applied with the Parwester, In terms of recommended field dose (RFD), Durban appeared almost 10 times more toxic than Sherpa: the 96-h LD50 for Durban was 0.057 (CI: 0.048-0.071) and 0.054 (CI: 0.046-0.066) RFD for spring and autumn beetles respectively, and for Sherpa ~ 0.556 (CI:0.453-0.704) and 1.003 (CI:0.863-1.214) RFD respectively. However, the toxicity of both insecticides was almost identical in terms of their active ingredients (g a.i. ha−1) – the LD50 for CPF was 16.4 for spring and 15.6 for autumn beetles, and for CYP 16.7 and 30.1, respectively. The beetles survival rate decreased significantly with increasing dose of both insecticides, but the spring-collected beetles appeared more sensitive, plausibly explained by their overwintering or ageing. In contrast to insectsicides, tebuconozol caused significant increase in survival at higher doses, possibly due to its interference with immune competence of insects or elimination of pathogenic fungi. The results show that at least some insecticide formulations may cause unacceptable effects on NTA when applied according to recommendations, indicating the urgent need for revising current pesticide usage recommendations. The differences in sensitivity between the spring and autumn-collected beetles call for further studies to see whether such seasonal differences can be important for ERA. This study was supported by National Science Centre, Poland (2015/19/B/NZ9/01939).

**TH146** A Field Trial to Determine Effects of Thiamethoxam treated Sugar Beet Seed on the Non-Target Arthropod Fauna of Arable Land in The Netherlands

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The aquatic environment is influenced by a broad range of chemicals and the toxic effects on species depend, amongst others, on the chemical properties of pesticides, soil, microbes, earthworms, plants... and these are poorly understood. Hence, in this contribution (poster), we would like to present the novel microcosm experiment, where the combined effects of soils properties, microorganisms, plants and earthworms on CF multimedia fate and bioavailability were investigated. In particular, the CF fate (by means of total, desorbable and freely dissolved concentrations) and bioavailability (by means of uptake to model fauna, flora and passive samples) is studied in complex microcosm systems consisting of agriculturally used fluviosols under the addition of selected model compounds (RECETOX); K. Brandstätter-Scherr, University of Natural Resources and Life Sciences / Institute for Environmental Biotechnology, Department for Agrobiotechnology (IFA-Tulln); J. Hofman, Masaryk University, RECETOX / Faculty of Science, RECETOX

The class of conazole fungicides (CFs), among them epoxiconazole, tebuconazole, flusilazole and prochloraz are currently used pesticides and members of the triazole group, used as broad-spectrum fungicides that inhibit ergosterol biosynthesis and are typically applied as foliar sprays for cereals, sugar beet or oilseed rape. 

Conazole fungicides are widely used in EU countries and their residues are frequently found in European arable soils which corresponds to their environmental properties. CFs are strongly sorbed to soil (log KOC of 3-4) and have low to moderate water solubility (SY of 7-150 mg/L). They are very persistent in soils and tend to form long-term residual conazole compounds in soil. CF fate (by means of total, desorbable and freely dissolved concentrations) and bioavailability will be investigated in a field study using the Kennard-Stone algorithm. These 10 soils are representative of a large fluviosol range in terms of their qualitative and quantitative SOM properties (TOC, DOC, HA, FA, %OC, pH, texture, etc.).
arthropod (NTA) populations, (2) the range of NTA taxa affected by the treatment and (3) the duration of treatment effects and the time period until populations recover. The evaluation will be based on (1) time to recovery (population density similar to control) and (2) persistence of effect (population growth similar to the control). Effects will be classified in accordance with De Jong et al. 2010.

**TH148 Bioaccumulation kinetics of pesticides chlorpyrifos and tefubonazole in the earthworm Eisenia andrei in two different soils**

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This study was conducted to investigate the bioaccumulation of two pesticides currently used in large amounts in the agriculture, the insecticide chlorpyrifos and the fungicide tefubonazole. The two compounds have distinct properties suggesting their different fate in the soil and bioaccumulation. Their detailed uptake kinetics in the model earthworm species *Eisenia andrei* were measured to find a sufficient length of exposure to achieve equilibrium in concentration between soil and earthworm and to compare two pesticides in two arable soils differing in the organic carbon (1.02 and 1.93% respectively) and clay content (10.7 and 20.7% respectively). Concentrations of the pesticides in soils and earthworms were determined by LC-MS/MS after QuEChERS extraction which has shown to be rapid, simple and effective approach to determine broad spectrum of pesticides in soil and earthworm samples. According to our results, a steady state was reached after 3 to 5 days for both pesticides and soils. The values of bioaccumulation factors calculated at the steady state ranged from 4.5–6.3 for chlorpyrifos and 2.2–13.1 for tefubonazole. Bioaccumulation factors were also calculated as the ratio of uptake at steady state to the concentration of the particular pesticide in the soil. The results suggested that the degradation and bioaccumulation of tested compounds by earthworms was influenced by more factors than only the organic carbon content in soils. The clay content also probably contributed, namely to degradation of chlorpyrifos by clay-catalysed hydrolysis and to decreased bioavailability of tefubonazole by binding to clay minerals. The lower Koc and hydrophobicity of tefubonazole relative to chlorpyrifos probably led to higher availability of tefubonazole through pore water exposure. On the other hand, higher hydrophobicity of chlorpyrifos probably caused an increase in availability by its additional uptake via ingestion.

**TH149 Effects of diuron and imidacloprid on eight nematode species**

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To assess the effect of two common herbicides, diuron and imidacloprid (insecticide) on ubiquitous organisms at the basis of food webs, we performed multispecies toxicity tests using nematode species commonly found in soil and freshwater benthic ecosystems. Diuron and imidacloprid belong to the top 15 of the most frequently detected pesticides in French rivers. Both chemicals show an elevated DT50 (time to 50% degradation) in sediments, about 130 days for imidacloprid and more than 300 days for diuron. In the current study, 10 representative species of different taxonomic groups, a representative for the response of nematode species to toxic stress, showed no significant response to imidacloprid at high concentrations up to 119 mg L−1 regarding growth or reproduction. Diuron inhibited 82% of reproduction success of *C. elegans*, but showed no significant effect on growth at high concentration (33 mg L−1). Then, we compared the lethal effects of diuron and imidacloprid on eight species of free-living nematodes: *Aphelenchoides sp, Caenorhabditis elegans, Pristionchus pacificus, Diplocapsus coronatus, Rhabditis sp, Plectus velox, Plectus opisthocirculus, Plectus acuminatus*. Nematodes were exposed in water for 48h to two concentrations (35 and 350 mg L−1) for imidacloprid and 10 and 100 mg L−1 for diuron). Results indicated a low risk of these pesticides to nematodes, as the chemicals did not affect significantly the survivorship at their solubility limit in water for every tested species.

**TH150 Multigeneration effects of pentachlorophenol and 2,2',4,4'-tetrabromodiphenyl ether on Folsomia candida**

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The multigeneration effects of pentachlorophenol (PCP) and 2,2',4,4'-tetrabromodiphenyl ether (BDE47) on the springtail Folsomia candida were evaluated. Multigeneration tests were performed in accordance to two different test methods. In the first method, the parental generation springtails (F0) were exposed to PCP or BDE47 for 28 days. The first filial generation (F1) springtails were transferred to unpolluted artificial soil for 28 days and reproduced the second filial generation (F2). In the second method, the F0 generation were exposed for 10 days and then transferred to unpolluted artificial soil to generate the F1 generation. The F1 generation were also transferred to unpolluted artificial soil for 28 days and reproduced the F2 generation. For PCP, significant effects were observed on F1 and F2 generation in the first method and F1 generation in the second method. This suggests that PCP influences the reproductive capacity of adult springtails and the hatching of eggs or the mortality of juveniles. For BDE47, significant effects were only observed on F1 generation in the first method, which shows that BDE47 affects egg hatching through the reproductive potential of adult springtails. Effects on the affected endpoints of springtails can be inferred by the two methods. PCP and BDE47 do not influence completely the same endpoints.

**TH151 Bioaccumulation of lead in earthworms: a comprehensive study to derive a bioaccumulation factor (BSAF) for risk assessment**

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Secondary poisoning to mammals and birds is a critical pathway for risk assessment of Pb in soil. This risk is generally assessed for the food-chain soil => earthworms => earthworm eating predators. Therefore, a correct evaluation of bioaccumulation of Pb in earthworms is essential for risk assessment of Pb in soils under the REACH Regulation and a literature review of biota-to-soil accumulation factors (BSAF) for Pb in earthworm species was made. To ensure that biota Pb burdens are in equilibrium with soil Pb concentrations, only data from field studies or laboratory studies using soil and biota collected at the same field site were considered. Data from laboratory studies where Pb was added to the soil as a Pb salt were only used as supporting evidence. In total, 248 BSAF values for earthworms were identified that meet the reliability criteria, ranging from 0.01 to 22.05 (dry weight basis, median 0.23). Results are available for several earthworm species, belonging to different ecological groups of earthworms: anecic, endogenic and epigeic earthworms. No distinct differences in BSAF values across these groups could be identified. BSAF values are derived in a wide range of soils and the data available can be considered as representing earthworms in Europe. Only the cation exchange capacity (CEC) is significantly correlated with BSAF values. No significant correlation with Pb content, pH, organic carbon content or clay content is observed. The significant negative regression between log BSAF and log CEC was confirmed by laboratory studies and is also consistent with the significant decrease in Pb toxicity observed for *Eisenia fetida* reproduction with increasing CEC of the soil. It was concluded to implement the effect of soil properties on BSAF by using the overall regression between log CEC and log BSAF in the risk assessment of Pb in soil. This yields a generic BSAF of 0.30 on dry weight basis, corresponding to 0.048 on a fresh weight basis, for the median eCEC value of 16 cmol/kg soil for European arable soils. BSAF values on fresh weight basis vary from 0.089 to 0.028 for soils with eCEC of 0.5 cmol/kg soil to 0.031 to 0.013 for 2 Me cmol/kg soil, respectively, corresponding to the 10th and 90th percentile of eCEC in European arable soils. Implementing effect of soil properties on BSAF improves consistency between assessment of secondary poisoning and direct toxicity of metals to soil organisms, where bioavailability corrections for varying soil properties are commonly accepted.

**TH152 Hazard assessment of liquid organic hydrogen carriers in terrestrial environment**

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A proactive environmental hazard assessment of liquid organic hydrogen carriers (LOHCs) – a novel energy system which can efficiently and reliably safely store and transport hydrogen – was conducted by characterisation of potential behaviours and ecotoxicities of these chemicals in soil environment. Adsorption properties of promising LOHC candidates including indoles, quinaldines, carbazole derivatives, benzyloleumnes and dibenzyleumnes in terms of carbon-water partition coefficients (Koc) were investigated via HPLC screening. Further characterisation was performed via adsorption isotherm modeling and soil column leaching with the extraction of soil-water by the liquid chromatography. The adsorption of Quin(2-Me) as examples. Soil ecotoxicity was estimated for the quinaldines in the soil bacteria *Aerobicacter globiformis* and Collemella Folsomia candida in pore-water and soil exposure scenarios. The log Koc values generally increased following indoles < quinaldines < carbazole derivatives < benzyloleumnes < dibenzyleumnes. The mobility of LOHCs was thus classified as highly mobile, moderately mobile or immobile. Adsorption isotherm and column leaching showed the strongest adsorption and retention of the partially hydrogenated form (Quin(2-Me)-ph) in soils. The H-rich form (Quin(2-Me-H10) appeared the highest leaching capacity through the soil followed by the H2-lean form (Quin(2-Me) implying the risk of groundwater contamination. Ion-interaction was considered dominant in the adsorption of Quin(2-Me-H10) to soils given its high protonation at the soil pH; while hydrophobicity was the main force in the adsorption of its two analogues. No or only slight toxicity was found for the quinaldines in the *Aerobicacter* at the highest test concentrations (500 mg L−1 and 750 mg kg−1 dry weight (dw) soil). Higher toxicity was found in the Collemella and mafollinations.
of cuticle in the pore-water scenario were observed. Dose-response modeling showed 10 < EC₅₀ < 100 mg L⁻¹ (liquid-only exposure) and 100 < EC₅₀ < 1000 mg kg⁻¹ dw soil (calculated soil pore-water based) of the quinaldines assigning these chemicals to category “harmful” to soil organisms. Predicted no-effect concentrations showed 1–3 orders of magnitude higher the effective concentrations than the former suggesting potential risks of the chemicals toward the soil environment and proper monitoring is needed in the application of the LOHCs. Key words: adorption, bioavailability, hazard assessment

TH153

Combining field measurements and biotest to assess lead and zinc phytoavailability in contaminated urban soils

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Along with the French legislation on the recycling of wastes from wastewater treatment plants, a guideline was provided to stakeholders to prove the low mobility and phytoavailability of trace elements exceeding total concentration thresholds in soil. Due to the lack of adequate plant biotest at the time the guideline was published (i.e. in 2005), the guideline suggests to measure trace element phytoavailability in the aerial parts of plants collected in situ in contaminated and uncontaminated soils. The present study aimed at applying the guideline methodology with the combination of a recently developed plant biotest (i.e. the RHIZOtest) and field measurements to lead (Pb) and zinc (Zn) contaminated urban soils on which irrigation with treated wastewater was foreseen. Ten contaminated and uncontaminated soil samples (hereafter referred to as soil) were collected in representative sites expected to be irrigated with treated wastewater. The phytoavailability of Pb and Zn was estimated on each soil by measuring Pb and Zn concentration in the aerial parts of field-collected plants and by deploying the RHIZOtest and measuring the uptake flux of Pb and Zn in the whole plants exposed to soil. As expected, field-collected plants exhibited a large range of Pb concentration in leaves, irrespective of total Pb and Zn concentrations and Pb and Zn mobility and phytoavailability measured in soils in the first step. In comparison, RHIZOtest measurements showed that only the contaminated soil 15 exhibited a significantly higher phytoavailability than other soils and have consequently to be represented for irrigation with wastewater. This study thus showed how the use of a biotest dedicated to the measurement of trace element phytoavailability in combination with field measurements was useful to assess the risk of high phytoavailability in contaminated urban soils.

TH154

Can approaches beyond the traditional ones characterize the effects on soil microflora provide an added value in the scope of regulation?


Aside from environmental regulation, side effects of chemical substances on the soil microflora focus on the determination of the nitrogen transformation (OECD 216). However, according to EFSAs more comprehensive risk assessment is required. We investigated whether a combination of several test approaches addressing various microbial aspects results in a better understanding of the fluctuation of the soil microflora after a threat and a more comprehensive risk assessment taking the necessary requirements into consideration. Specifically, we aimed to consider ecosystem services and the protection of the biodiversity into account. We used a silver nanomaterial as test for irrigation with wastewater. This study thus showed how the use of a biotest dedicated to the measurement of trace element phytoavailability in combination with field measurements was useful to assess the risk of high phytoavailability in contaminated urban soils.

TH155

Re-calibration of the earthworm Tier 1 risk assessment of plant protection products - an update

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The determination in tier 1 of the appropriate plant protection products (PPP) is expected to increase due to revision of the PECbio modeling guidance. The new EFSA guidance foresees to use worst case PECbio values for each European regulatory zone considering a lower soil bulk density, a lower organic carbon content, and a reduced crop interception rate due to consideration of worst case wash-off assumptions. Furthermore, several different soil layers for which PECbio values could be calculated are under discussion, i.e. 0-1 cm, 0-2.5 cm, 0-5 cm, and 0-20 cm soil depth. Calculated PECbio values based on the new EFSA guidance are estimated to strongly increase, which might lead to an overly conservative tier 1 risk assessment. In a project published by Christl et al. (2016), results from standard laboratory earthworm reproduction tests are compared with the effect levels in higher tier-studies for a representative active of 5 soil samples supplied by ECPA companies. In this exercise, the relevant soil layer for PECbio modeling and assessment factor were adjusted to result in tier 1 protection of unacceptable field study effects. The results of this evaluation give clear evidence that considering a layer of 0-5 cm in combination with the currently used assessment factor of 5 would lead to an appropriate earthworm tier 1 risk assessment (Christl et al. 2016). Concerning the lower tier, the target is to establish a tool to assess earthworm toxicity and interspecies variability by taking the deviation of laboratory endpoints regarding correction of laboratory endpoints for lipophilic compounds (logP > 2). A correction of endpoints by a factor of 2 is proposed by EFSA (2015, EFSA Supporting publication 2015:EN-924) for studies containing artificial soil with 5% peat (formerly only endpoints from studies with 10% peat were corrected for high logP). Furthermore, in its Scientific Opinion, EFSA (2017) proposed Specific Protection Goals for earthworms which include a maximum acceptable recovery time of 6 months for initial effects in field studies. This deviates from the current procedure of an acceptable recovery time of one year for earthworm populations. The dataset of 54 case studies was re-evaluated considering the new EFSA proposals and the new results will be presented.

TH156

Digging into the soil risk assessment of pesticides: current approach and its uncertainty

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According to the Uniform Principles (Reg No 546/2011) in the context of Regulation (EC) No 1107/2009, all possible sources of uncertainties should be considered when performing a regulatory assessment in the context of pesticides authorization. The current risk assessment for soil organisms, conducted according to SANCO/10329/2002, foresees, at Tier 1, the application of a trigger value of 5. A missing uncertainty lineart trend in the interaction and interspecies variability in the extrapolation of toxicity endpoints from lab- to field. However, the current approach presents additional uncertainties. Test methodology for soil organisms only requires dosing verification after the application of the pesticide to the soil. The determination of the tested concentration at regular intervals is currently not required although it may be very relevant for a proper hazard characterization (e.g. bioavailability), since, for example, during laboratory handling procedures of the spiked soils, possible losses of the pesticide may occur. In case further refinements of the risk are triggered, higher tier tests (semi-field or field studies) under more realistic conditions may be one option. Standardised field protocols are mainly available for earthworms. The available standardised field methods evaluate the effects on abundance and biodiversity of earthworms, taking into consideration the likely level of effects, the species/groups affected, population recovery (within 1 year) as well as information on the application and fate of the pesticide. The magnitude of effects is directly assessed in terms of risk without the application of any assessment factor. However, field studies only give a picture of a particular situation as effect manifestation and recovery are dynamic processes which depend on the local situation and time-scale. Additionally, an assessment at community level which takes into account interspecies interactions and indirect effects is currently not implemented as well as the statistical power of test is not properly evaluated. An approach aimed at defining a Regulatory Acceptable Concentration (RAC) could be useful to address those uncertainties and would allow the harmonisation of the risk assessment of the different taxonomic groups.

TH157

SETAC Soils Interest Group

M.H. Wagelmanns, Bioclear earth

Natural toxins and harmful algal blooms (HABs): water and food safety, analysis, toxicity, and risks (P)
A novel analytical method for simultaneous quantification of Bracken fern produced carcinogenic ptaquiloside-like compounds and their derivatives

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Significant variety of toxic secondary metabolites produced by plants appear in chemical structures of glycosidic molecules. These compounds are highly water soluble and mobile in soils and sediments. In cases when toxic glycosides are persistent and released in high loads from vegetation, evaluation of the risk to aquatic environments adjacent to drinking water supplies is needed. Nevertheless, the detection limits of individual and fate of these two toxins are largely unknown. This study aims to identify the environmental risk factors that predominately release of glycosidic natural toxins from non-agricultural lands to aquatic ecosystems. Bracken ferns (Pteridium aquilinum) are known to produce up to 6 kg/ha of carcinogen ptaquiloside. Previous studies demonstrate leaching of ptaquiloside from Bracken to soils and upper ground waters. The ptaquiloside-like compounds – ptaquesulfoside and caudatoside – have recently been studied in Australian Brackens. Except from a few positive samples included in the Australian study, there have been no reports of these compounds in Europe. We hereby report a novel method for quantification of ptaquiloside, caudatoside and ptaquesulfoside and their respective pterosin-derivatives (6 compounds in total) to be used for the aforementioned study. The novel LC/MS method (Agilent 1260 Infinity HPLC/MS) suitable nature. Ensuring sample integrity for analysis is a critical step to facilitate trustful findings, and appropriate preservation methods need to be developed. This research focuses on the development of a preservation technique for ptaquiloside (PTA) and its degradation product pteroquinobin (PTB) in groundwater. Ptaquiloside is a carcinogenic compound produced by one of the five most common plants on the planet, Bracken fern (Pteridium aquilinum). It is highly water-soluble with almost no retention to soil and sediment, and hence leaches to the aqueous environment. In turn, PTA can potentially contaminate groundwater, which presents a concern for human health if used as a drinking water source. Ptaquiloside is chemically unstable under acidic and alkaline conditions, making it difficult to collect and preserve for analysis. Thus, a controlled and well-designed preservation procedure that can ensure its stability for the subsequent analyses is necessary. In order to develop a technique for preservation of PTA and PTB in groundwater samples, a Plackett-Burman experimental design is applied. This approach allows assessing the influence of a number of independent factors such as sample bottle type, test time, water type, pH, temperature and transportation conditions by a reduced number of experiments. In each of the experiments, a water sample with known concentration of PTA and PTB was treated in a preservation kit. The recovery percentage of the compounds were evaluated by LC-MS system. This led to an optimal combination of factors for the preservation of the compounds of interest. We also performed robustness and range tests to quantify the precision, accuracy and linearity of the method. The optimized technique was further validated by applying it at field sites covering different groundwater types and different spiked toxin concentrations. By using this method, facilitated reliable investigation and monitoring of PTA and PTB in groundwater. In that way, we contribute both to the scientific discourse on the topic as well as offer a practical tool for water supply companies. This research project is part of European Training Network – NaToAxq, which is funded by the European Union’s Horizon 2020 research and innovation programme under the Marie Skłodowska-Curie grant agreement No. 722493.
easily computed for classes of compounds. The most significant factor was extraction temperature, especially for volatile early-eluting compounds where fine-tuning of temperature is essential to achieve the required sensitivity. The optimized automated HS-SPME-GC/MS method is proved to be a valuable tool for high-throughput, efficient and sensitive non-targeted screening of HAB odorous compounds, while sensitivity is further enhanced when certain classes of compounds are targeted, e.g. when sensory pre-evaluation of samples is applied. Acknowledgments: The authors thank CYANOCOST – COST Action ES 1105 www.cyanocost.net

TH163
Suspected screening of cyanotoxins in freshwater by high performance liquid chromatography coupled to high-resolution mass spectrometry D. Filatova, IDAEA- CSIC / IDAEA; m. picardo, IDAEA CSIC Barcelona / IDAEA; O. Núñez, Universitat de Barcelona / Department of Chemical Engineering and Analytical Chemistry; M. Farre, IDAEA-CSIC / Environmental Chemistry

Cyanobacteria are one of the components of the microbial assemblages in peri-dynamics. The community distribution is affected by water quality, flow regime, climate, and geology. During the past decades, there has been a noticeable increase of cyanobacterial blooms, dominating in many freshwater bodies worldwide. Some cyanobacteria species are known to produce toxic secondary metabolites called cyanotoxins, which vary in structure and harmful properties (hepatotoxins, neurotoxins), and being a major concern for drinking water supply and recreational water use. The most widespread cyanotoxins are microcystins (MCs) variants MC-LR, -RR, -YR, with MC-LR being the most toxic one. For this reason, the World Health Organization appointed a guideline of 1 µg/L in drinking water for total MC-LR. In order to monitor levels of cyanotoxins and prevent both human poisoning and wildlife damage, suitable analytical methods need to be developed. This work presents the development of a sensitive, fast and robust method for the analysis of cyanotoxins on high-performance liquid chromatography coupled to high-resolution mass spectrometry (HPLC-HRMS). For the sample pre-treatment, solid-phase extraction for multiple toxins has been employed, which was recently developed in our research group. The chromatographic separation was achieved using a C18 analytical column (150x2.1 mm, 5µm) using methanol and water as mobile phase. The total chromatographic run was 15 min. The chromatographic separation was coupled to a Q-Exactive Orbitrap instrument (Thermo Fisher Scientific). The interphase used was ESI under positive conditions. The main advantage of high-resolution mass spectrometry will be the target analysis of 10 cyanotoxins, as well as the analysis in scan mass spectrometry to assess the potential presence of transformation products and other non-targeted toxins in the samples. This multi-toxin method has been developed and validated for freshwater cyanobacteria such as microcystins, nodularin, cylindrospermopsin, and anatoxin-a. The developed method was applied for the study and characterization of cyanotoxins concentrations in Catalonia freshwater reservoirs. Keywords: cyanotoxins, microcystins, high-resolution mass spectrometry. M. Farre has recently received a Ph.D. grant from the European Union’s Horizon 2020 research and innovation programme under the Marie Skłodowska-Curie grant agreement No. 722493.

TH164
Oligonucleotide probes for fluorescence in-situ identification of cyanobacterial cells in surface waters L. deiana, Water Research Institute National Research Council / National Water Research Institute; A. Barra Caraciolo, National Research Council / Water Research Institute; P. Gennari, National Research Council of Italy (CNR) / Water Research Institute; c. fajardo, Faculdad de Veterinaria, Complutense University Avenida Puerta de Hierro s/n, 28040 Madrid, Spain; M. Martin-Fernandez, UCM / Biochemistry and Molecular Biology; l. medlin, Marine Biological Association of the UK, The Citadel, Plymouth PL1 2PB, UK; G. Mengers, Natural Biotec SL; m. saccli, Council for Agricultural Research and Economics (CREA), Agriculture and Environment Research Center (AA), Via di Corticella 133, 40128 Bologna, Italy; m. lettieri, European Commission, DG Joint Research Centre, Directorate D Sustainable Resource Recovery Water and Marine Resources TP 121, Via E.Fermi, 2749, 21027 Ipru (VA), Italy. Harmful cyanobacterial blooms have been increasing in freshwater ecosystems in recent decades, mainly because of eutrophication and climate change. In some cases, some cyanobacterial species can produce toxins and this phenomenon can have a negative impact and pose a risk for ecosystem and human health. Of the 15 known cyanobacteria genera, more than 40 species produce toxins, which are natural compounds showing different chemical and toxicological characteristics. Cyanobacterial toxins are responsible for both acute and chronic poisoning in animals and humans. Among the main classes of cyanotoxins, microcystins are among the most frequently found in the environment. These toxins are accumulated mainly in the liver, but also in the intestines and kidneys and can be very dangerous for both animal and human health (Lucernini and Ottaviani, 2011). Fast and sensitive methods to identify unequivocally Microcystis aeruginosa and Planktothrix agardhii are very useful to discriminate these species with respect to the other non-toxic cyanobacteria. For this purpose, we designed, developed and validated some oligonucleotide probes (GNPlankS02, PKAgD03, MicAerD03) for FISH (Fluorescence In-Situ Hybridization) analysis to detect these species in freshwater samples. The FISH probes were designed using the ARB software with the Silva database in the framework of the MicroCoKit project. We tested different fixedative methods to minimise the natural autofluorescence from chlorophyll-a (Groben and Medlin, 2005) to visualize Microcystis aeruginosa and Planktothrix agardhii under a laser confocal microscope. Firstly, the FISH probes designed have been tested on pure cultures of M. aeruginosa and P. agardhii species, then the probes were successfully applied to natural samples collected from surface waters. Keywords: Microcystis aeruginosa; Planktothrix agardhii; FISH probes; algae bloom. References Groben R. and Medlin L., 2005. In situ hybridization of phycoplancton using fluorescently labeled RNA probes. Methods in enzymology. 392:301-310, Eawag, CH. TH165
Adquacy of EPI Suite prediction models to estimate physicochemical properties of natural toxins potentially present in surface water J. Rodríguez Leal, Stockholm University / Department of Environmental Science and Analytical Chemistry ACES; M. MacLeod, ITM - Stockholm University / Department of Environmental Science and Analytical Chemistry

Natural toxins constitute a potential risk to water supplies in Europe. Only a few risk assessment models exist regarding different natural toxins. One of the main gaps is that production and occurrence of natural toxins are not extensively studied but not fully assessed (Bucheli 2014). Persistence and mobility of natural toxins might be usefully modelled in screening applications using techniques developed for environmental pollutants of anthropogenic origin, such as EPI Suite™ (US EPA 2017). Environmentally relevant partitioning properties of many natural toxins have not been experimentally determined. To model overall persistence of natural toxins in aquatic environments requires sorption coefficients (e.g., Kd) and estimates of their degradation rate constants in the aquatic environment that have been determined by experimental methods or estimated using quantitative structure-activity relationship (QSAR) and quantitative structure-property relationship (QSPR) models. QSAR predictions should be considered carefully when applied to a set of chemicals that are structurally distinct from those that were used to develop the model. The establishment of an applicability domain of the models provides a range of chemicals where the predictions are expected to be reliable and based on interpolation rather than extrapolation, regarding the structure of the chemicals in the training set (Gramatica 2007). We present here an analysis of the applicability domain of selected EPI Suite™ QSAR models, and interpret the results with reference to natural toxins within these limits that could be included in a deterministic prioritization of natural toxins in water according to their persistence and mobility. References Bucheli, Thomas D. 2014. “Phytoxins: Environmental Micropollutants of Concern?” Environmental Science & Technology 48 (22):13027–33. Gramatica, Paola. 2007. “Principles of QSAR Models Validation: Internal and External.” QSAR & Combinatorial Science 26 (5):694–701. US EPA. 2017. Estimation Programs Interface Suite™ for Microsoft® Windows, v 4.11. United States Environmental Protection Agency, Washington, DC, USA.

TH166
Cyanobacterial oligopeptides of environmental concern and (co)production dynamics R. Sanches Natumi, E. Vorwyl, Eawag Swiss federal Institute of Aquatic Science and Technology / Department for Environmental Chemistry, E. M. Janssen, Eawag Swiss federal Institute of Aquatic Science and Technology / Department of Environmental Chemistry

Our ecosystems and drinking water resources are not only vulnerable towards anthropogenic pollutants. Natural toxins present an additional threat for which we need comprehensive risk assessment. Cyanobacteria are known to produce a range of chemicals where the predictions are expected to be reliable and based on interpolation rather than extrapolation, regarding the structure of the chemicals in the training set (Gramatica 2007). We present here an analysis of the applicability domain of selected EPI Suite™ QSAR models, and interpret the results with reference to natural toxins within these limits that could be included in a deterministic prioritization of natural toxins in water according to their persistence and mobility. References Bucheli, Thomas D. 2014. “Phytoxins: Environmental Micropollutants of Concern?” Environmental Science & Technology 48 (22):13027–33. Gramatica, Paola. 2007. “Principles of QSAR Models Validation: Internal and External.” QSAR & Combinatorial Science 26 (5):694–701. US EPA. 2017. Estimation Programs Interface Suite™ for Microsoft® Windows, v 4.11. United States Environmental Protection Agency, Washington, DC, USA.
peptide abundance throughout the growth phase of single strains (e.g., Microcystis aeruginosa and Anabaena flos-aquae) and under different culturing conditions (e.g., N:P ratios and light intensities) by multifactorial analysis. New insights into co-production dynamics offer critical information about cyanotoxin mixtures present during harmful algae blooms and with that critical knowledge towards comprehensive risk assessment.

TH167
Degradation of the carcinogenic ptaquiloside under alkaline conditions
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The carcinogenic ptaquiloside (PTA) is found in several species of ferns worldwide. The distribution and occurrence is well described for genus Pteridium (Bracken fern). PTA is present in all parts of the fern. PTA is suspected of causing Human gastric cancer. PTA is a non-sesquiterpene glycoside and is not sorbed by soils to a great extent (logKow of approx. -0.5). Hence, PTA can leach from Bracken stands. Leaching is most prominent during rain but baseline levels are found in streams in Bracken infested areas. Soil contamination and contamination of upper aquifers has been observed on a number of occasions. PTA may contaminate groundwater resources. Dissipation of PTA under environmental conditions is governed by a number of factors: Enzymatic activity; pH (hydrolysis); irreversible sorption/reactions; and sorption to clay minerals. Bacterial activity and hydrolysis are the most important mechanisms causing dissipation of PTA. The purpose of this study was to describe the underlying mechanisms for the hydrolysis of PTA and formation of reaction products under near-sterile alkaline conditions as found in calcareous aquifers. PTA (4,700ppb) was deglycosidated using 0.010/10.0 M NaOH and 3 different 0.025M buffer systems (approx. pH 7-12, NaHPO4/NaH2PO4/H2BO3, pH regulated with 0.1M NaOH). Dissipation of PTA and formation of reaction products were monitored up to 200hrs at 25°C. PTA and the main reaction product pterosin B (PTB) was quantified by LC-MS using SIM- and TIC-modes. Formation of other reaction products was analysed semi-quantitatively using the relative areas of the main mass traces. Chemical rate constants are reported for degradation of PTA and species formation. Dissipation of PTA were pH dependent as previously observed. Addition of 0.01-1.0M NaOH results in immediate degradation of PTA and formation of a small amount of PTB plus two more reaction products. The concentration of PTB remained constant over time. PTB is the endproduct of hydrolysis in pure solutions. One reaction product was identified as the bracken dieneone (BDE), a ultimate carcinogen. BDE is formed immediately while the unknown is formed from BDE reaching stoichiometric balance within the timeframe of the experiments. Rate of formation were dependent on pH, as well as of the buffer system used. The results indicate that PTA under alkaline conditions will form carcinogenic reaction products that are stable in groundwater.

TH168
Experimental Determination of Octanol-Water Partitioning Coefficients of Natural Toxins
C.D. Schoenesee, Agroscope / Environmental Analytics; K. McNeill, ETH Zurich / Institute of Ecological Chemistry and Pollutant Dynamics; T. Bucheli, Agroscope ART / Environmental Analytics
The octanol-water partitioning coefficient (Kow) remains one of the key parameters in environmental fate and risk assessment of organic chemicals for regulatory purposes. Based on predicted Kow values, many natural toxins are expected to be mobile in the aquatic environment. If such toxins are not retained well in soils, they may not be effectively removed by water resources treatment. For compounds such as natural toxins, with physicochemical and structural complexity due to large numbers of various functional groups, current estimation models for Kow, and other phase distribution coefficients show limited applicability. Thus, experimentally determined physicochemical property data are still of great value to regulatory organizations defining thresholds for potential environmental contaminants. In this study, Kow values are experimentally evaluated by both indirect and direct approaches based on well-established OECD methods modified for application in natural toxin analysis. A multi-step molecular size and functional group specific calibration procedure is applied in indirect evaluation of natural toxin partition properties based on chromatographic retention. Direct analysis of partitioning behavior is performed in a miniaturized shake flask system in standard HPLC vials. As HPLC based methods, both approaches show the capability to be largely automated for more efficient, less error-prone analysis and thus allow the reliable determination of Kow in the for potential aquatic contaminants relevant range of log Kow from -2 to 5. Natural toxins for analysis comprise previously investigated mycotoxins and isoalloxazines as reference compounds in addition to representatives of specific plant classes. In regards to predicted toxicity, persistence and mobility as well as plant occurrence, specific natural toxin subclasses such as pyrrolizidine alkaloids from Senecio spp. or quinolizidine alkaloids from Lupinus spp. are investigated in more detail. As an indicator for the partitioning of natural toxins from aqueous media to organic matrices, Kow can be seen as first proxy estimating natural toxin mobility in the aquatic environment. Thus, experimental data will help in prioritizing of toxins for further research activities, including field studies and lab-based characterization of fate processes e.g., within the current MC-ITN NaToxAg. [1] ECTOC; Technical Report No. 123, 2013 [2] Schenzel, et al.; Environ Sci Technol2012.
Cyanobacteria form blooms in freshwaters due to environmental pollution and can cause toxic effects. A novel treatment method involves the use of the bacterium Pseudomonas putida PAO1, which can degrade toxic compounds from cyanobacterial blooms. This approach is promising for the removal of cyanotoxins from drinking water, as traditional drinking water treatment methods may not be sufficient. Advanced Oxidation Processes (AOPs) are emerging treatment methods that have shown promise for the removal of organic pollutants, including cyanotoxins. AOPs promote the in situ formation of highly reactive radicals, which can break down complex molecules such as cyanotoxins. The study describes the use of AOPs for the removal of cyanotoxins from drinking water, highlighting the potential for these technologies in addressing the growing threat of cyanotoxin-contaminated water sources. The research contributes to the development of more effective treatment methods for aquatic ecosystems, aiming to protect public health and the environment.
B1 (AFB1) and total aflatoxin (AFT), as a screening test, was used in order to analyze imported nuts, from non-European countries, intended for direct human consumption. The percentage of AFS positive samples (only pistachios and almonds), taken during the three years from 2013 to 2015, under the national programs of official control, amounted to 9% for B1 and 10.5% for AFT, and were confirmed by HPLC (High Performance Liquid Chromatography). The results demonstrated that pistachios from Turkey were contaminated by AFS but very rarely in freshwater organisms. Bivalves are known to consume bioaccumulation of BMAA has recently been demonstrated with highly selective neurodegenerative disease amyotrophic lateral sclerosis (ALS), is less studied. The Interest of bivalves for the biosurvey of cyanotoxins in aquatic ecosystems to a synergistic effect on DNA damage implying possible consequences for the mussel populations. The higher incidence of AFS in imported shelled nuts from Turkey are more broadly contaminated and confirmed by HPLC (High Performance Liquid Chromatography). The results demonstrated that pistachios from Turkey were contaminated by AFS but very rarely in freshwater organisms. Bivalves are known to consume bioaccumulation of BMAA has recently been demonstrated with highly selective bioaccumulation of the hepatotoxins microcystin (MC) and anatoxin-a, in the samples from the Czech Republic. Levels of tetrodotoxin and anhydro-tetrodotoxin. Agric. Biol. Chem. 50:793–795. [3] Kodama M, Sato S, Sakamoto S, Ogata T. 1996. Occurrence of tetrodotoxin in ovaries of puffer fish Fugu rubripes. Toxicon 45:851–854. [4] Yamasu T, Yasumura D, Yotsu M, Michishita T, Endo A, Kotak Y. 1986. Bacterial production of tetrodotoxin and anhydro-tetrodotoxin. Agric. Biol. Chem. 50:793–795. [5] Kodama M, Sato S, Sakamoto S, Ogata T. 1996. Occurrence of tetrodotoxin in Alexandrium tamarense, a causative dinoflagellate of paralytic shellfish poisoning. Toxicon. 34:1101–1105. [6] TH178 Tetrodotoxin an Emerging Threat to Humans in the Mediterranean Area: First Detection in Italian Mussels C. Dell’Aversano, University of Napoli Federico II, Department of Pharmacy / Pharmacy; L. Tartaglione, F. Varriale, University of Napoli Federico II / Department of Pharmacy; A. Penna, University of Urbino / Department of Biomedical Sciences; M. Giacobbe, Institute for Coastal Marine Environment, CNR; S. Piggozo, A. Milandrini, Fondazione Centro Ricerche Marine, P. Bordin, L. Biglia, Istituto Zooprofilattico delle Venezie; A. Turner, Plymouth University / Food Safety. Tetrodotoxin (TTX) is one of the most potent neurotoxins, originally found in ovary and liver of pufferfish (Tetrodontidae) [1]. Successively, TTX was isolated from other marine and terrestrial animals, as xanthid crab, trumpet shellfish, blue-ringed octopus, gastropods, starfish, and frogs. The wide distribution of TTX in genetically unrelated organisms has made TTX origin for long time controversial, with different kind of bacteria being identified as TTX-producing organisms [2]. Even Alexandrium tamarense – one of the paralytic shellfish poisoning toxins (PST) producing organisms – was proposed as potential biogenic source of TTX [3]. Although fatal human poisonings following consumption of TTX-contaminated seafood have been reported so far only in Japan, the accumulation of TTX in fish, oysters and mussels collected in Europe (Spain, Portugal, UK, Greece) has been recently reported. So, in the frame of a collaborative study on evaluation of PST-related risk in the Mediterranean area, mussels collected in the Siracuse bay (Sicily, Italy) over a three year period (2015-2017), were analyzed by hydrophilic interaction liquid chromatography coupled with both high resolution and tandem mass spectrometry detection (HILIC-HRMS and HILIC-MS/MS). Both techniques highlighted the presence of high PST contamination levels, with samples collected in 2016 containing up to 10851 µg STX eq/kg. Unexpectedly, together with PST, tetrodotoxin was detected in Sicilian mussels. Although this was the first report of TTX in Italy, contamination levels found in mussels (0.8-6.4 µg/kg) were well below the regulatory limit of 2 mg. TTX eq/kg established for TTX in Japan. Interestingly, much higher contamination levels of TTX (413 µg/kg) have been detected in mussels collected in 2017 in the NE Adriatic coasts of Italy (Lagoon of Marano), in the frame of the monitoring programme for marine biotoxins regulated in the EU. [1] Wu Z, Xie L, Xia G, Zhang J, Nie Y, Hu J, Wang S, Zhang R. 2005. A new tetrodotoxin-producing actinomycete, Norcardiopsis dassonvillei, isolated from the marine sponge Pachymastus sp. Toxicon 45:851–859. [2] Yamasu T, Yasumura D, Yotsu M, Michishita T, Endo A, Kotak Y. 1986. Bacterial production of tetrodotoxin and anhydro-tetrodotoxin. Agric. Biol. Chem. 50:793–795. [3] Kodama M, Sato S, Sakamoto S, Ogata T. 1996. Occurrence of tetrodotoxin in Alexandrium tamarense, a causative dinoflagellate of paralytic shellfish poisoning. Toxicon. 34:1101–1105. [4] TH177 The first report on neurotoxic anatoxin-a occurrence in cyanobacterial blooms in the Czech Republic L. Blahova, Research Centre for Toxic Compounds in the Environment (RECETOX) / A. Hilscherova, Masaryk University, Faculty of Science, RECETOX / Research Centre for Toxic Compounds in the Environment RECETOX: O. Lepova Skacelova, Faculty of Science, University of South Bohemia; L. Blaha, Masaryk University, Faculty of Science / Research centre for toxic compounds in the environment RECETOX. Prototypical cyanotoxins such as microcystins have been extensively studied all around the world but there is still a lack of research on the occurrence levels and risks of other toxic metabolites produced in harmful blooms of cyanobacteria. In this paper we present the results of the first survey focusing on less explored cyanotoxins, namely anatoxin-a, in the samples from the Czech Republic. Levels of cyanobacteria were analyzed in freeze-dried biomass collected during 2012-2015 in various reservoirs in the country. The focus was on blooms (total 34 samples) dominated by potential producers of anatoxin-a such as Dolichospermum sp. (syn. Anabaena sp.), Aphanizomenon sp. as well as blooms formed by less common cyanobacteria. The multi-target UPLC-MS/MS methodology was applied that allowed to analyze in parallel all major cyanobacterial toxins (microcystin-LR,
incidents in freshwaters and in finis
Cyanobacteria as well as eukariotic algae produce a wide range of volatile
Hiskia, NCSR Democritos; S. Triantis, NCSR Demokritos / Institute of Nanoscience and Nanotechnology; A. Diaz
Cyanobacteria taste and odor compounds; a study in freshwaters of Greece
TH181
triggering the blooms, as well as a
m. Cyanobacteria are not always present (from Nov 2016 until Jul 2017 no species
cell/L, respectively). Disueri Lake is among the largest lakes, with a surface of 1.85
composition of the cyanobacterial community changed dramatically. In July
population and wild and domestic animals. Between mid
summer. After the July sampling when a
physical, microbiological and microscopic) analysis of samples, according to the
targeted and non-targeted analysis based on automated HS-SPME-GC/MS for fast
Samples of lakes and water reservoirs of Greece were collected for T&O analysis
beyond geosmin and MIB. Acknowledgement - The authors thank CYANOCOST
COST Action ES 1105 www.cyanocost.net. C. Christophorides acknowledges the
program of Industrial Scholarships of Stavros Niarchos Foundation

TH182
Determination of multi-class cyanotoxins in fish tissues
C. Christophoridis, National Center for Scientific Research / Institute of
Nanoscience and Nanotechnology; I. Argyropoulos, NCSR Demokritos / Institute
of Nanoscience and Nanotechnology; C. Christophoridis acknowledges the
COST Action ES 1105 "CYANOCOST" and the program of Industrial Scholarships of Stavros Niarchos Foundation
especially FAs have great structural diversity and high biological specificity, essential for every living cell, as sources of energy, as membrane constituents, or as metabolic and signaling mediators. FAs have long been used as food-web tracers, and, more recently, changes in FA profile have also been exploited to better understand how contaminants affect organisms in aquatic food-webs (Silva et al. 2017). In this study, the potential impact of A. armata exudates in the FA profile of two marine invertebrates was assessed. For this, after calculating the lethal concentrations of the alga exude, *Gibbula umbilicalis* and *Palauophora serratus* were exposed for 168 hours to non-lethal concentrations of this exudate. Consequently, the trends in changes of FA abundance and composition were evaluated separately in the body tissue of the snails and in the shrimps hepatopancreas. Results showed different FA profiles between invertebrates but for both species the profile was influenced by exudate concentration exposure, with significant alterations being observed for several saturated FA and polyunsaturated FA like Acid Arachidonic (ARA), EPA or DHA. These alterations can represent an impact on these organisms’ cell function since some of these FA have important communication and signaling roles within and between cells. Fatty acid profile showed to be a sensitive and highly informative parameter to address effects of marine organisms, both predator and prey in cohabiting invertebrates, thus constituting a promising tool for understanding biological pollution effect assessment in these coastal organisms.

**TH184**

**Impacts of Asparagopsis armata on marine invertebrates: behavioral and biochemical responses**

C.O. Silva, Polytechnic Institute of Leiria; C.E. Silva, S.C. Novaís, Polytechnic Institute of Leiria / MARE IPILeiria; M.F. Lemos, Instituto Politécnico de Leiria / MARE IPILeiria

The introduction of non-native seaweeds outside their native distributional range, through human activities, has been causing documented negative effect on native species. The red algae *Asparagopsis armata*, with its invasive strategy including specialized cells capable of producing compounds with potent biological effects, is capable of inducing significant changes in terms of native community composition. This is specially occurring in rocky pools (intertidal zone) where *A. armata* releases several compounds that in these enclosed and extreme conditions are often adverse for other organisms such as other seaweed, vertebrates, and invertebrates leading to severe consequences for coastal ecosystems. The main objective of this study was to evaluate the potential impact of *A. armata* on marine invertebrates by exposing the common prawn *Palaeomon serratus* and the marine snail *Gibbula umbilicalis* to the exudate of this macroalga. The seaweed collected at the coast of Peniche, Portugal was left in laboratory tanks, for 12 hours, in the dark at 20°C±1. After assessing the lethal concentrations of the algae exude, animals were exposed for 168 hours to non-lethal concentrations of this exudate and subsequently analyzed for biochemical biomarkers responses associated with detoxification (glutathione S-transferase, GST), antioxidant defenses (catalase, CAT; superoxide dismutase, SOD), oxidative damage (lipid peroxidation, LPO; DNA damage), neurotoxicity (acetylcholinesterase, AChE) and energy metabolism (lactate dehydrogenase, LDH; Isocitrate dehydrogenase, IDH; electron transport system activity, ETS; content in lipids, proteins and carbohydrates). Also, behavioral endpoints were performed using the flipping behavior for *G. umbilicalis* and the avoidance behavior for *P. serratus*. The biomarker responses analysed on invertebrates showed an impairment of their physiological status after exposure to this algae exudate, with both species suffering from disruptions in their neuronal and energy metabolism functions, as well as from oxidative stress inducing damage in their macromolecules. Behavioural responses were also highly sensitive to *Asparagopsis* exudate exposure. These results represent an important step in the research of toxic exudates released to the environment and can serve as warning indicators of prospective effects of this macroalga on the invasive ecosystems under a global change scenario.

**TH185**

**Assessing consumption risks through cadmium-contaminated shellfish amplified by ocean acidification**

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The purpose of this study is to assess the human health risk of Taiwan population through consumption of cadmium-contaminated hard clam and oyster amplified by ocean acidification. This study employed forecasted ocean surface pH from the coupled global model integrated with projected emission scenarios representative concentration pathways 8.5, and C distribution as 0.001 – 2 μg L⁻¹ in Taiwan coast to estimate potential Cd accumulation of shellfish. A gender-specific physiologically-based pharmacokinetic model was developed to assess urinary and blood Cd concentration via daily shellfish consumption. The dose-response function was used to account for the prevalence of renal dysfunction and osteoporosis in response to human accumulated Cd in urine and blood. Results showed that median Cd accumulations under current and 2100 ocean acidification scenario were, respectively, 0.0009 and 0.0010 μg g⁻¹ for hard clam, whereas 0.0186 and 0.0210 μg g⁻¹ were estimated for the oyster. The urinary Cd concentration in female had potential 80% higher for ocean acidification scenario than that for non-ocean acidification. However, results revealed that exceedance risks of renal dysfunction and osteoporosis for gender-specific consumption only and whole groups under 2100 ocean acidification were no significant difference with that of current non-ocean acidification scenario. This study concluded that ocean acidification was not likely to increase synergistically the renal dysfunction and osteoporosis of human health risk through shellfish consumption.

**TH186**

**Cyanobacterial toxins - a threat to the human respiratory tract?**

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Intensified occurrence of toxic cyanobacterial blooms is becoming a major human health threat. Microcystin-LR (MCLR) is probably the most frequent and abundant cyanobacterial toxin known to humans. However, the metabolic and signaling mediators. FAs have long been used as food sources of energy, as membrane constituents, or as signaling molecules to address effects of marine organisms, both predator and prey in cohabiting invertebrates, thus constituting a promising tool for understanding biological pollution effect assessment in these coastal organisms. In this study, the potential impact of *A. armata* exudates in the FA profile of two marine invertebrates was assessed. For this, after calculating the lethal concentrations of the alga exude, *G. umbilicalis* and *P. serratus* were exposed for 168 hours to non-lethal concentrations of this exudate. Consequently, the trends in changes of FA abundance and composition were evaluated separately in the body tissue of the snails and in the shrimps hepatopancreas. Results showed different FA profiles between invertebrates but for both species the profile was influenced by exudate concentration exposure, with significant alterations being observed for several saturated FA and polyunsaturated FA like Acid Arachidonic (ARA), EPA or DHA. These alterations can represent an impact on these organisms’ cell function since some of these FA have important communication and signaling roles within and between cells. Fatty acid profile showed to be a sensitive and highly informative parameter to address effects of marine organisms, both predator and prey in cohabiting invertebrates, thus constituting a promising tool for understanding biological pollution effect assessment in these coastal organisms.

**TH187**

**Effects of microcystin-LR and cyanobacterial LPS in human airway in vitro models**

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Intensified occurrence of toxic cyanobacterial blooms is becoming a major human health threat. Microcystin-LR (MCLR) is probably the most frequent and abundant cyanobacterial toxin detected in the environment, known to induce primarily hepatotoxic effects. Therefore, in this study, we investigated the toxicological effect of MCLR in human airway epithelial cells and the marine snail *Gibbula umbilicalis* to the exudate of this macroalga. The seaweed collected at the coast of Peniche, Portugal was left in laboratory tanks, for 12 hours, in the dark at 20°C±1. After assessing the lethal concentrations of the algae exude, animals were exposed for 168 hours to non-lethal concentrations of this exudate and subsequently analyzed for biochemical biomarkers responses associated with detoxification (glutathione S-transferase, GST), antioxidant defenses (catalase, CAT; superoxide dismutase, SOD), oxidative damage (lipid peroxidation, LPO; DNA damage), neurotoxicity (acetylcholinesterase, AChE) and energy metabolism (lactate dehydrogenase, LDH; Isocitrate dehydrogenase, IDH; electron transport system activity, ETS; content in lipids, proteins and carbohydrates). Also, behavioral endpoints were performed using the flipping behavior for *G. umbilicalis* and the avoidance behavior for *P. serratus*. The biomarker responses analysed on invertebrates showed an impairment of their physiological status after exposure to this algae exudate, with both species suffering from disruptions in their neuronal and energy metabolism functions, as well as from oxidative stress inducing damage in their macromolecules. Behavioural responses were also highly sensitive to *Asparagopsis* exudate exposure. These results represent an important step in the research of toxic exudates released to the environment and can serve as warning indicators of prospective effects of this macroalga on the invasive ecosystems under a global change scenario.
exposures to LPS (10 μg/mL) also did not significantly decrease cell viability and neither MCLR nor LPS affected gap junctional intercellular communication in bronchial cell lines. Regardless of MCLR uptake, the toxin was relatively less cytotoxic to human bronchial epithelial cells when compared to the effects of other cyanotoxins (e.g. cylindrospermopsin), or in comparison with other cell types (e.g. hepatic or neural cells). Further experiments should focus on more detailed characterization of MCLR uptake and on long-term effects of MCLR and LPS on inflammation-related endpoints. Inhibition toxicity of other harmful cyanobacterial metabolites could include compounds with estrogenic and/or retinoid-like activity. Endocrine disruptive compounds can cause disturbance in hormone signalling and adverse effects in biota even at very low concentrations. Retinoids play a role as important signalling molecules which control vital cell processes like morphogenesis, development, reproduction or apoptosis. This study focused on freshwater ponds and reservoirs affected by water blooms and determined the estrogenic and retinoid-like activity of water by in vitro bioassay as well as concentrations of main estrogenic and retinoid compounds by LC-MS/MS analyses. Water samples from freshwater reservoirs and ponds with water blooms elicited estrogenic activity up to 2 ngEQ/L. This activity could be only partly explained by the concentrations of analysed estrogenic hormones, alkylpheno derivatives and endocrine disruptors. Other compounds might play a role in these effects as well. Maximal detected retinoid-like activity in water samples reached 256 ng EQ/L. We analysed the presence of nine retinoid substances, where 4-keto all trans retinoic acid and retinal were the most common forms detected in the samples. The activity was almost fully explained based on concentration and retinoid-like activity in water samples collected during peak bloom conditions. Current results show that different cladocerans had different sensitivities to microcystin. In this study we conducted with several different cladoceran species using lyophilized phytoplankton samples. Herrera, Echeverri and Ferrao (2015) also investigated in the future. The research was supported by the Czech Science Foundation Grant No. GJ17-23279Y and H2020-MSCA-ITN-2016 Project No.722493 NTeQox.

TH188

Estrogentic and retinoid-like activity in stagnant waters M. Smutna, Masaryk University, Faculty of Science, RECETOX / RECETOX Research Centre for Toxic Compounds in the Environment; T. Prochazkova, Research Centre for Toxic Compounds in the Environment, Faculty of Science, Masaryk University / Research centre for toxic compounds in the environment; J. Priebojová, Masaryk University, Faculty of Science, RECETOX / Research centre for toxic compounds in the environment RECETOX; E. Sychrová, Masaryk University, Faculty of Science, RECETOX / RECETOX Research Centre for Toxic Compounds in the Environment; L. Sehnal, Masaryk University Faculty of Science RECETOX / RECETOX Research Centre for Toxic Compounds in the Environment; K. Hilscherová, Masaryk University, Faculty of Science, RECETOX / Research Centre for Toxic Compounds in the Environment RECETOX Cyanobacterial water blooms represent worldwide problem in many freshwater as well as marine ecosystems as producer of substantial amount of various bioactive compounds, some of which can cause adverse effects and pose risk to both aquatic organisms and humans. Recent investigations indicate that cyanobacterial metabolites could include compounds with estrogenic and/or retinoid-like activity. Endocrine disruptive compounds can cause disturbance in hormone signalling and adverse effects in biota even at very low concentrations. Retinoids play a role as important signalling molecules which control vital cell processes like morphogenesis, development, reproduction or apoptosis. This study focused on freshwater ponds and reservoirs affected by water blooms and determined the estrogenic and retinoid-like activity of water by in vitro bioassay as well as concentrations of main estrogenic and retinoid compounds by LC-MS/MS analyses. Water samples from freshwater reservoirs and ponds with water blooms elicited estrogenic activity up to 2 ngEQ/L. This activity could be only partly explained by the concentrations of analysed estrogenic hormones, alkylpheno derivatives and endocrine disruptors. Other compounds might play a role in these effects as well. Maximal detected retinoid-like activity in water samples reached 256 ng EQ/L. We analysed the presence of nine retinoid substances, where 4-keto all trans retinoic acid and retinal were the most common forms detected in the samples. The activity was almost fully explained based on concentration and retinoid-like activity in water samples collected during peak bloom conditions. Current results show that different cladocerans had different sensitivities to microcystin. In this study we conducted with several different cladoceran species using lyophilized phytoplankton samples. Herrera, Echeverri and Ferrao (2015) also investigated in the future.

TH189

Excitatory effects of 2,4 -diaminobutyric acid on leech Retzius nerve cell membrane potential S. Spánik, Faculty of Medicine, University of Belgrade / Institute for Pathophysiology; M. Stanojević, Faculty of Medicine, University of Belgrade / Institute for Pathophysiology Ljubodrag Buba Mihailović; M. Prostran, Faculty of Medicine, University of Belgrade / Institute for Pharmacology, Clinical Pharmacology and Toxicology; S. Lopičić, Faculty of Medicine, University of Belgrade / Institute for Pharmacology Ljubodrag Buba Mihailović; M. Smutna, Faculty of Medicine, University of Belgrade / Institute for Pathophysiology Ljubodrag Buba Mihailović; R. Feiters, University of Veterinary Medicine, Austria; J. Allen, U.S. EPA / Office of Research and Development; T. Sanan, U.S. EPA / Office of Research and Development; J. Allen, U.S. EPA / Office of Research and Development; J. Allen, U.S. EPA / Office of Research and Development; J. Allen, U.S. EPA / Office of Research and Development; J. Allen, U.S. EPA / Office of Research and Development; J. Allen, U.S. EPA / Office of Research and Development; J. Allen, U.S. EPA / Office of Research and Development; J. Allen, U.S. EPA / Office of Research and Development; J. Allen, U.S. EPA / Office of Research and Development. Literature indicates that LC50 for microcystin-RR in rainbow trout is ~21 μg/L. There is even less ecotoxicity information available for prymnesin which is produced from the estuarine algae Prymnesium parvum. This flagged alga has invaded freshwater systems in the U.S. and has caused numerous fish kills recorded in inland Texas lakes and blooms in 10 other states. Given the uncertainty with the purity of existing toxin standards and the cost of using them to conduct toxicity studies, a new approach is proposed using pure cultures and ambient bloom samples. Herrera, Echeverri and Ferrao-Filho (2015) conducted toxicity tests on several different cladoceran species using lyophilized photopankton samples collected from hydroelectric/dinking water reservoirs in Brazil. They found that reservoir samples with higher microcystin contents were the most toxic ones and that different cladocerans had different sensitivities to microcystin. In this study we have taken a similar approach but have used laboratory cultures of a toxin-producing strain of unicellular Microcystis aeruginosa, non-toxic producing filamentous strain of Anabaena flos-aquae and P. parvum. Each culture was centrifuged to separate cells from their respective culture media, then resuspended in moderately hard water. The M aeruginosa cells were then frozen/thawed 3 times at -80°C. The flos-aquae cells were then freeze-dried. Forty-eight-hour acute assays were conducted with Ceriodaphnia dubia, Hyalella azteca larval Pimephales promelas and Neocloeon triangulifer on both strains. A similar procedure was also used on lake water samples collected during peak bloom conditions. Current results show microcystin concentrations of 74 μg/L did not cause any significant acute toxicity to any of the four test species. The filamentous non-toxic producer A flos-aquae caused significant mortality to N triangulifer and H azteca only when tested in moderately hard Reconstituted Water (only when tested in Moderately Hard Reconstituted Water) which are both grazer feeders. Mechanism of effect is undetermined at this time. August 2017 Lake Harsha bloom sample (300,000 cells/mL > WHO high risk probability value) was not acutely toxic to any of the 4 test species. Additional P. parvum acute results and microcrystin chronic results will also be presented.

TH190

Proteomic analysis of rice plant exposed to long-term microcystin-LR exposure Xiaoying Nanjing Institute of Environmental Sciences, MEP Irrigation with cyanobacterial-blooming water containing microcystins (MCs) poses potential threat to the growth of agricultural plants. Rice (Oryza sativa L.) is an important grain crop and is widely grown for domestic consumption in China. However, large amounts of rice field in the middle part of China has been irrigating with cyanobacterial-blooming water. So far, the molecular mechanism of MC induced inhibition of cell proliferation and apoptosis of rice cells remains unclear. In the present study, rice plants were exposed to 1.0 μg/L and 50 μg/L of microcystin-LR (MC-LR) in the hydroponic nutrient solution for 34 days. The proteomic profiles of rice leaves after exposure were analysed using tandem-mass-tag labelling and LC-MS/MS analysis. The results showed that a total of 298 differentially expressed proteins were found, 89 differentially expressed proteins of them in 1.0 μg/L MC-LR treatment group, and 289 differentially expressed proteins in 50.0 μg/L MC-LR treatment group. Different response characteristics of protein expression were found in rice leaves exposed to low-concentration (1.0 μg/L) and high concentration (50.0 μg/L) of MC-LR,
respectively, and the different biological pathways involved in the mechanism of MC-LR-induced toxicity to rice were revealed using GO Term and KEGG analysis. Exposure to 1.0 μg/L and 50 μg/L of MC-LR could disturb the photosynthetic and ribosome pathways in rice leaves, causing the adverse effects on the normal growth and photosynthesis of rice. The significant alterations of the biological processes induced by the exposure to 50 μg/L of MC-LR were the inhibition of ribosome, porphyrin and chlorophyll metabolism, photosynthesis and terpenoid backbone biosynthesis related pathways, and the induction of thioredoxin, inositol phosphate metabolism, vitamin B6 metabolism and flavonoid biosynthesis related pathways in rice leaves. These results provided evidence that the molecular mechanisms underlying adverse effects in terrestrial plants exposed to water containing microcystins (MCs).

Keywords: rice, microcystin-LR, photosynthesis, proteomics

Acknowledgments
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Developments in the use of bioassays for chemical and environmental risk assessment (P)

TH114 Responses to PFOA and PFBS exposure in the sediment dwelling invertebrate Dreidboaena veneta (Annelida)

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The aim of this work was to evaluate the neurotoxicity of Polyfluorinated alkyl substances (PFASs) in the species Dreissena polymorpha, a freshwater mussel species that accumulates PFASs in its tissues. The effects of PFASs on the invertebrate, Dreidboaena veneta (Annelida), bioaccumulation patterns and cellular and biochemical responses in coelomocytes (metastasis and lyososomal membrane stability), and at tissue level (GPX and MTS), following the exposure to two perfluorinated alkyl acids (PFOA and PFBS) for short (72 h) and longer (14 and 28 days) times. The exposures were carried out in a microcosm prepared with glass containers filled with 300 ml of soil humified at 30% with PFOA or PFBS spiked water.

As for the 72 h tests the exposures were performed, at two different nominal concentrations, i.e. 30% of 1x or 10x MAC-EQS fw values (Maximum Acceptable Concentration-EQS calculated by the Italian Working Group for the derivation of Environmental Quality Standard (EQS)) while for the long time the PFOA, or PFBS, nominal concentrations were 30% of 5x MAC-EQS fw values. Different accumulation patterns were observed for PFOA and PFBS, with PFOA no longer accumulating between 14 and 28 days, while PFBS continues to be accumulated up to 28 days. Significantly higher coelomocyte mortalities than in the controls, with both compounds, were detected after the 14 and 28 days exposures. As for the lysosomal membrane stability significant decreases were detected both after the short and the long-time exposures. In the soft tissues primary data don't show significant differences between control and treated organisms regarding the GPX activity. A significant MTS total decrease was detected after PFOA exposure, both at 14 and 28 days while after PFBS exposure only at 14 days. As for MT, because it has been reported that PFAFs seem to increase ROS levels, we determined not only the total protein concentration but also the oxidized fraction (MTox). A significant increase in the MTox fraction in PFOA treatment after 28 days and in PFBS after 14 days was observed. On our results show, for this invertebrate organism, a higher PFBS bioaccumulation than PFOA and significant exposure effects to the two PFASs both in coelomocytes, the main immunodefensive system cells of the organism, and in the soft tissues. Further studies are planned to explore the mechanisms underlying these results.

TH115 Toxicity of Per- and Polyfluoroalkyl substances on Chironomus dilutus for use in a relative toxicity model

C.J. McCarthy, CH2M / Environmental Services; M. Stanley, B. Muckey, Test America; C. Salice, Towson University / Environmental Science & Science Dept.; D. Wright, CH2M

Per- and polyfluoroalkyl substances (PFASs), including perfluorooctanoic sulfonate (PFOS) and perfluorooctanoate (PFOA) are commonly Elevated in soil and groundwater. High detection frequency and concentration has resulted in identification of PFASs as compounds of interest and as emerging contaminants due to their regulatory uncertainty. Published toxicological research to date relates to PFOS and PFOA only and for a limited number of organisms. The lack of robust and defensible ecotoxicity data on other PFASs hinders risk assessment and leads to unsupported risk management decisions. Given this gap in understanding of the additional compounds, the Strategic Environmental Research and Development Program (SERDP) is funding research of these additional PFASs and classes of organisms. This discussion will summarize the first phase of a SERDP research grant to address these needs. Tests were conducted with a common aquatic test species to identify patterns of relative toxicity between the PFASs. Chironomus dilatus tests included a 96-hour reference toxicant test, a 10-day range finding test, and a 20-day definitive bioassay. For shorter duration Chironomus tests, the main endpoint of interest was survival while for longer-duration tests (20 days), the more sensitive growth endpoint was measured. Opportunistic measurements of defensibility were also included to enhance the understanding of potential toxicity to these compounds. Test results will ultimately be used in conjunction with concurrent tests being conducted with avian and reptilian model species to the same chemicals to develop a relative toxicity model. Endpoints measures from the aquatic species tests will be used to identify clear patterns of relative toxicity of the tested PFASs. Results will inform and prioritize PFAS testing on avian and reptilian model species. In addition, the relative potency patterns observed after aquatic testing will be reassessed upon completion of the upper trophic level exposure studies. Once all phases of testing are complete, the results will be used to help develop a
risk management framework for addressing potential environmental management issues of PFAS.

TH196 Interpretation of bioassay results in the context of the soil quality TRIAD approach. N. Pandur, INERIS; S. ANDRES, INERIS / Toxicological Ecotoxicological Assessment of chemical Substances; P. Pandur, INERIS / Expertise and assay in ecotoxicology unit

The recently standardized method ISO 19204 “Soil quality – Procedure for site specific ecological risk assessment of soil contamination (Soil quality TRIAD approach)” describes in a general way the application of three combined lines of evidence (chemistry, ecotoxicology and ecology) along a tiered approach. Regarding the ecotoxicological component, the TRIAD approach consists in carrying out a battery of bioassays on soil samples and to scale the results from all bioassays to calculate an ecotoxicological combined risk score. In order to evaluate its applicability and the relevance of the proposed tools, INERIS used the soil quality TRIAD approach on an applied case: an open mine operated for 60 years and in which the acid generation stopped about 10 years ago, in the vicinity of a river replicate. The results indicated both species- and strain-related differences in binding affinities of these ligands, and suggest there is potential for these exogenous chemicals to interact and possibly influence vertebrate thyroid hormone-dependent function.

TH197 Estimating the hazardous concentrations of nonylphenol for soil ecosystem protection with presence approach. J. Kaak, J. Mood, D. Kim, Konkuk University; R. Cui, Konkuk University / Department of Environmental Sciences; Y. An, Konkuk University / Department of Environmental Health Science

Nonylphenol is known as a xenoestrogen but is still used for pesticides, detergents, surfactant cleaners and packaging. However, few researches on soil ecotoxicity of nonylphenol were reported. In the present study, we conducted a battery of bioassay, generated soil ecotoxicity data and then estimated soil hazardous concentration for nonylphenol for soil ecosystem protection based on species sensitivity distribution (SSD). For the acute assay, eight soil species from six different taxonomic groups (magnoliopsida, liliopsida, chlorophyceae, clitellata and collembo) were tested. Also, for the chronic assay, five soil species from four different taxonomic groups (magnoliopsida, liliopsida, chlorophyceae and collembo) were investigated. Finally, acute and chronic hazardous concentrations for HCl, HCO3, H2SO4 were suggested for protection of soil ecosystem. These values can be applied to environmental risk assessments for nonylphenols. This study was funded by the Korea Ministry of Environment (MOE) as the Environmental Health Action Program (1485014458).

TH198 Organophosphate Triesters and Selected Metabolites Enhance the Binding of Thyroxine to Human Transthyretin In Vitro K.L. Hill, Instrinski / Department of Biology; R.J. Letcher, Environment and Climate Canada / Ecotoxicology and Wildlife Health Division; T. Hamers, Wageningen University, Institute for Environmental Studies (IVM) / Department of Environment and Health; J. Kamstra, NMBU / BaSam; W. Wilmore, Carleton University / Department of Biology

The toxicological properties of organophosphate (OP) triesters that are used as flame retardant and plasticizer additives are currently not well understood. Furthermore, as increasing the concentration for nonylphenol for soil ecosystem protection based on species sensitivity distribution (SSD), the results indicated both species- and strain-related differences in binding affinities of these ligands, and suggest there is potential for these exogenous chemicals to interact and possibly influence vertebrate thyroid hormone-dependent function.

TH200 Phosphine changes cytochrome c oxidase in Sitophilus oryzae K. Kim, H. Kim, H. Jeon, Y. Choi, Y. Kim, S. Lee, Kyungpook National University

Phosphine resistance in the stored product insect pests has been reported over the world. In South Korea, Sitophilus oryzae has been developed phosphine resistance after the severe use of phosphine. In this study, how S. oryzae survived under the recommended dose of phosphine and we assessed the biochemical and molecular mechanisms for referring phosphine resistance. S. oryzae were prepared as control groups (C), medium-resistant group (MR), and strong resistant groups (R) for this study. One target sites of phosphine is cytochrome c oxidase (COX) and we analyzed the enzyme activities within the three strains. The highest COX activities were found in R groups with about 1.5-fold increase when compared to the controls. IC50 values on the COX activity by ethyl formate, one of COX inhibitors was 2.82, 3.71 and 5.85 mM for C, MR, and R strains. Lineweaver-Burk plot for COX using ethyl formate exhibited different modes from R strains to C strains. And six genes related to COX enzyme and regulation of phosphine resistance in S. oryzae may be related to the changes in COX enzyme and up-regulation of phosphine gene expressing juvenile hormone indecipherable protein. Three strains of S. oryzae were chosen for the experiment and the results were analyzed using RT-PCR for comparing gene expression and cat gene was dramatically down-regulated in the R strain. Phosphine induced COX enzyme expression of the gene was also up-regulated in the MR strain, but it was not so big different. Three biomarker enzymes such as acetylcholinesterase, glutathione S-transferase, and carboxylesterase activities were also determined within the three strains. Only glutathione S-transferase activity decreased in the R strain. Taken together, phosphine resistance in S. oryzae may be related to the changes in COX enzyme and up-regulation of phosphine gene expressing juvenile hormone inducible protein.

TH201 Effects of additives in mobile phases in simultaneous analysis of glutathione and glutathione disulfide by HPLC-MS/MS S. Berek, KIST Europe / Environmental Safety Group; Y. Jung, KIST-Europe; Y. Kim, KIST Europe / Environmental Safety Group

Glutathione is an important non-protein compound and existed in both internal and external cells. Regarding toxicological effects induced by oxidative stress, ratio of reduced form (GSH) to oxidized form (GSSG) of glutathione is one of important biomarkers. Among all available assays to detect and quantify GSH and GSSG, using high-performance liquid chromatography (HPLC) coupled with mass spectrometry (MS) is essential with the development of high sensitive and sensitive analytical methods. GSH and GSSG are usually analyzed in positive ionization in electrospray, and formic acid can be a general additive in mobile phases for a better protonation in positive ionization mode in MS. In this work, we investigated the effects of additives other than formic acid for a better understanding to enhance the ionization of GSH and GSSG in the gas field of MS source. With the presence of formic acid only, as a result, protonation of GSSG was very limited. However, using formic acid and ammonium acetate together in mobile phases delivered the enhancement of protonation for both GSH and GSSG. Furthermore, as increasing the concentration of ammonium acetate from 5 mM to
50 mM, sensitivities of GSH and GSSG were increased from 0.0034 to 0.0072 and 0.028 to 0.046, respectively. Detection limits of both GSH and GSSG were also significantly lowered as using higher concentration of ammonium acetate. This indicates that producing ammonium adducts followed by dissociating ammonium ions from adducts to protonate GSH and GSSG is important mechanism for protonation of these compounds with using ammonium acetate as a mobile phase additive. This enhanced sensitivity was also used for identification and quantification of both GSH and GSSG and achieved more than 100% recovery for GSH and around 100% recovery for GSSG. The achievement of higher recovery of GSH than 100% was because ZPL itself might not be oxidized to form GSSG. In addition, same method was also applied to ZPL exposed to different concentrations of a target chemical as well as 6 mg/L of H2O2, a negative control. The lowest concentration of GSH in this work was 5.0 ng/ml, higher than its detection limit, 2.0 ng/ml. This is meaningful because it could not be achieved by other conventional methods and assays with higher detection limit than its original concentration. Therefore, we can conclude that our method could avoid underestimation to quantify biomarkers such as GSH and GSSG.

TH202 Rapid analysis of bivalves’ xenobiotabolome using High Resolution Mass Spectrometry

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A mixture of xenobiotics is released to the environment every day from residential, commercial and industrial uses. They are simultaneously present at different levels in aquatic ecosystems making a “cocktail” of hazardous substances. These xenobiotics interact with wild organisms and may be bioaccumulated. They can have negative implications from an environmental point of view, affecting wild life, but also they may be of great concern from a human health perspective, when they accumulate in highly consumed organisms like bivalves. Given that it is unrealistic to assess every possible combination of chemical substances accumulated by organisms, the major challenge now is to develop systematic ways of addressing these chemical mixtures, and to identify priority mixtures of potential concern. For this purpose, the profiling of the xenobiotabolome, or range of xenobiotics and their metabolites in an organism exposed to environmental contaminants, seems to be the way forward. In the present work, for the profiling of the xenobiotabolome, a fast analytical method has been developed for the extraction and identification of priority contaminants in bivalves from Ebro Delta, Spain. A literature research was done in order to gather all the information available regarding the Ebro Delta and possible sources of contamination. Taking into account the information a mixture of compounds representative of the different contaminant sources identified in the area was selected. This mixture including pesticides, plasticizers, antibacterials, preservatives, stimulants, and pharmaceuticals was used for a recovery study with three different bivalves types of high commercial interest such as mussel, oyster and cockle. QuEChERS (Quick, Easy, Cheap, Effective, Rugged and Safe) was used for the extraction and purification of the pesticides. The purifying extracts were injected in Orbitrap-Q-Exactive for identification and quantification of the priority contaminants. Twenty compounds including endocrine disrupting compounds, pesticides, and pharmaceuticals were extracted with recoveries ranging from 40.54 to 105.51 %. Quality parameters such as method detection and quantification limits, accuracy, and precision were studied. Besides, non-target analysis of other relevant contaminants that may be present in bivalves’ xenobiotabolome is ongoing.

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TH203 River ecosystem: an ecosystem approach to evaluate the ecological risk linked to the human health protection

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The anthropogenic pressure on river ecosystems can induce changes on their structural and functional characters as well as an increasing risk for human health. Over the last years an ecosystem approach mainly based on multilevel bioindicator methods has been developed for assessing the risk for human health. The samples processing plays a key role in the environmental analysis. Samplings were carried out in an area characterized by strong anthropogenic pressures (Tiber River Central Italy). The water samples were processed using the technique of filtration and concentration, as suggested within the European project μAQUA (2015), at the same time, tests were carried out using raw water samples. The approach of this study is based on the following parameters: biological community (diatoms, macro invertebrates, macrophytes and fishes fauna); chemical–physical parameters, a set of ecotoxicological bioassays (Vibrio Fischeri, Daphnia magna and Vicia faba), microbiological analysis (Salmonella spp, Staphylococcus spp, Clostridium spp, and Campylobacter upland virological analysis including in vivo Hemolytic activity, HEV and HEV, Norovirus NoGI and NoGiL, Reovirus, Enterovirus: A, B and C, Adenovirus: ADV40 and ADV41). The results of this study showed that the pre-concentration of larger amount of water improves both the contaminants detection within aquatic ecosystems and the ecological risk evaluation. The ecotoxicological analysis as part of the integrated approach to evaluate the ecological risk linked to the human health protection. The integrated approach adopted has been a useful tool to describe the ecological status of surface waters and the related risk for human wellbeing, providing a complete and organic vision of the qualitative state of the ecosystem. In conclusion these results highlighted the different levels of alteration and the potential need for the primary prevention and restoration, confirming that an ecosystem approach plays a key role in the ecological and human health risk assessments.

TH204 INTEGRATED EXPOSURE AND EFFECT DATABASE TOOLS TO SUPPORT HAZARD AND RISK ASSESSMENT


Use of available exposure and effect data are key to performing hazard and risk assessment of pollutants, and compiling different sources of data are often done in a case-by-case manner. Processing data is thus cumbersome and time consuming, whereas the availability of data is a large source of uncertainty in resulting assessments. The NIVA Risk Assessment database (NIVA RArdSM) has been developed as a module-based tool to facilitate the assembly, organisation, integration, visualisation and quality assurance of available exposure and effect information in order to speed up and perform consistent handling of relevant data. The NIVA RArdSM compile available experimental and predicted (computational) effect data that range from molecular and cellular responses characterising the mode of action (MOA), typically derived from high-throughput and high-content (in vitro) bioassays, to (apical) adverse data derived from whole organism bioassays of potential regulatory relevance. These effect data are assembled within the context of Adverse Outcome Pathways (AOPs) hierarchically organized into a cellular responses referred to as molecular initiating events (MIE), to downstream key events (KE) at the cellular/organ level and finally to adverse outcomes (AO) at the individual or organism level. The resulting multi-level assemblies of data can be used in hazard assessment to identify the MOA of one or more stressors, to link molecular responses to higher organisation level effects and to identify potential stressors among large assemblies of pollutant risks that can give rise to a given AO. The NIVA RArdSM also support risk assessment by calculating risk quotients (RQs) of single pollutants and mixtures of these on basis of exposure (typically measured or predicted environmental concentrations) and effect data (typically NOEC, ECx, PNEC or EQS values) and can identify risk drivers (most toxic chemicals), relevant toxic endpoints (i.e. MIE, KE and AO) and susceptible species for a given exposure scenario. Recent developments include integration of non-chemical stressors such as ionizing and non-ionizing radiation. Examples on uses specific exposure scenarios will be presented to show the utility of the databases and the tools developed. Acknowledgments: RCN projects 221455-EDRISK (www.niva.no/edrisk), 268294 MixRisk (www.niva.no/mixrisk), 223268 CERAD (www.mmbu.no/en/services/centers/cerad), and EU-FP7 project SOLUTIONS (http://www.solutions-project.eu/project/).

TH205 Assessing exposure risk for marine bivalve Mytilus posed by microplastic polystyrene particles

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BACKGROUND: Microplastics (MPs) are one of the most widespread emerging pollutants in aquatic ecosystems, posing impacts on marine organisms. However, little is explored for potential risks of environmentally relevant concentrations of MPs or MPs based on bioassay results from related published studies. Recent development includes integration of non-chemical stressors such as ionizing and non-ionizing radiation. Exams on use specific exposure scenarios will be presented to show the utility of the databases and the tools developed. Acknowledgments: RCN projects 221455-EDRISK (www.niva.no/edrisk), 268294 MixRisk (www.niva.no/mixrisk), 223268 CERAD (www.mmbu.no/en/services/centers/cerad), and EU-FP7 project SOLUTIONS (http://www.solutions-project.eu/project/).

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RESULTS: We found that PNECs for inhibition of lysosomal membrane stability and phagocytic cells were 0.04 and 0.07 μg mL\(^{-1}\), respectively, implicating that phagocytosis is a more sensitive endpoint for immune responses in bivalves. In addition, our results demonstrated that the North Pacific Ocean appeared to be the greatest risks among global oceans. CONCLUSIONS: The strict thresholds estimated by applying a environmental risk assessment framework could be recommended as a criteria for environmental management of PS-MPs or MPs. Potential effects of PS-MPs/MPs on marine organisms at higher trophic levels should also be taken into consideration. Keywords: Polystyrene microplastics; Bivalve; Environmental risk assessment; Predicted no-effect concentration; Predicted environmental concentration; Hazard quotient

TH207
Innovative Design of Nationwide Dutch Water Quality Monitoring
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According to the European Union Water Framework Directive (EU-WFD), chemical surface water quality is assessed by analysing the concentration of 45 priority compounds. However, the analysed chemicals are often absent and biological effects are thus caused by (unknown) mixtures of compounds. Alternatively, water quality can be assessed by observing adverse effects of surface water on test organisms. Therefore, the present study aimed to improve surface water quality assessment by applying an innovative design in a nationwide monitoring campaign in The Netherlands. To this purpose bioassays with two aquatic invertebrate species, Daphnia magna and Chironomus riparius, were employed and the performance of passive sampling techniques to include time integrated compound concentrations was explored. D. magna neonates and C. riparius larvae were exposed to surface water samples from 34 locations. Daphnids were additionally exposed to POCIS passive sampler extracts from 7 of these locations. For the daphnids, none of the surface water samples or passive sampler extracts caused significant mortality after 48h of exposure. In contrast, for the chironomids, three surface water samples caused significantly lower larval survival compared to the controls. The use of C. riparius bioassays thus allowed for differentiation between water quality of the sampling locations. Possible explanations for the observed chironomid mortality include insecticide sorption to the provided food, which may lead to increased exposure resulting in higher mortality. A possible culprit compound could be the neonicotinoid imidacloprid, which was detected at two locations with observed chironomid mortality. Moreover, toxicity of imidacloprid to C. riparius is 500 times higher than to D. magna. This could thus explain the high mortality at these greenhouse locations. It is noted that using bioassays with multiple test species provides better insight into surface water quality, and is therefore a valuable addition to regular water quality monitoring.

TH208
Smart Monitoring: Application of innovative tools in nationwide water quality assessment
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The European Union Water Framework Directive requires member states to assess chemical water quality of surface waters by monitoring the presence of 45 priority substances. However, these substances are often banned and their concentration in surface waters is strongly decreased, frequently to levels below the limits of detection. Simultaneously, industries have switched to a myriad of alternative substances. However, these substances are often not listed as priority compounds, but meanwhile could prevent costly chemical analysis at sites with low ecotoxicological risks.

TH209
Passive sampling in effect-based monitoring of two European rivers - expressability of in vitro effects of detected chemicals
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The use of passive sampling as a tool in environmental monitoring has gained wide acceptance. Thus, passive sampling and use of effect-based tools in the monitoring of aquatic pollution, has combined approaches was used for monitoring of two rivers differing significantly in pollution levels. The Bosna, moderate-sized river in Bosnia-Herzegovina, which is burdened by untreated wastewaters, was sampled by semipermeable passive sampling devices (SPMD) and POCIS samplers. The Danube, the largest river in the EU with relatively low pollution level, was sampled using a mobile dynamic passive sampling device with silicone rubber (SR) and SDB-RPS Empréteur(TM) (ED) disc samplers. Both sampler sets consisted of partitioning sampler for non-polar chemicals (SPMD, SR) and adsorption sampler for the polar-ones (POCIS, ED). For the partitioning samplers, concentrations of collected chemicals in river water were derived using dissipation of performance reference compounds. For the adsorption samplers, the sampling rates were either taken from the literature (POCIS) or calculated from the chemical concentrations that were detected both in adsorption (ED) and partitioning samplers (SR). The samples were analyzed for aryl hydrocarbon-, estrogen- and androgen receptor-mediated effects using in vitro bioassays. The effects were expressed as bioanalytical equivalents (BEQs) of respective model compounds in water. The BEQs were specifically calculated in extracts from POCIS and ED samplers showing that the polar chemicals were responsible for most of the detected effects. Chemical analyses detected 103 and 209 chemicals in the Bosna and the Danube samples, respectively. The passive sampling allowed detection of chemicals at pg/L concentrations. The levels of chemicals with known biological potency for the studied endpoints were used for modeling of bioanalytical equivalents (BEQ\(_{\text{bio}}\)). The use of bioanalytical equivalents showed that the detected chemicals explained mostly a low fraction of the BEQ\(_{\text{bio}}\). Only in the case of estrogenicity in extracts of the samplers collecting polar chemicals, the BEQ\(_{\text{bio}}\) was comparable with the BEQ\(_{\text{chem}}\) levels. Both sampler combinations proved to be suitable for the detection of a large set of chemicals even at trace levels and for the complementary assessment of the biological potentials of the environmental mixtures. The SOLUTIONS Project was supported by the 7th Framework Programme EU (FP7-ENV-2013) with grant agreement no. 603437.

TH210
Testing of realistic contaminant mixtures with the harpacticoid copepod species Nitocra spinipes using passive sampler extracts
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The use of passive sampling as a tool in environmental monitoring has gained wide acceptance within the past decades. More recently the possibilities of combining passive sampling and biotesting gained higher attention and researchers focus on reconstituting environmentally realistic contaminant mixtures in aquatic biotest systems. Equilibrium based samplers (e.g. silicone rubber sheets) can mostly be used as passive dosing devices in biotest systems without prior treatment but have the disadvantage that only one single concentration level can be tested. For equilibrium samplers (e.g. Speedisks(TM)) an extraction is needed before spiking of the biotic medium and the downside of this approach is that an extraction always changes the natural mixture composition due to compound specific partition coefficients. The advantage on the other hand is that the extraction of the samplers is well established, efficient and easy to combine with chemical analysis. Thus, even though this approach does not reconstitute the natural concentration profile quantitatively it allows for a comprehensive in silico and in vitro based monitoring of two environmental samples to derive ecotoxicologically relevant concentrations in mixtures in terms of qualitative chemical composition. In the recent study we extracted Speedisk™ passive samplers that were deployed for 8 weeks in two Belgian harbours and one location next to one of the harbours to spike a 7-day larval development test with the harpacticoid copepod Nitocra spinipes following ISO 18220. In order to fractionate the compounds on the Speedisks™ we followed two different procedures: a sequential and a parallel extraction approach using three solvents: acetonitrile, ethyl acetate and dichloromethane. We exposed 80 larval megalops to 8 replicates in a fully randomized setup including controls and solvent controls to each of the Speedisk™ extracts and counted larvae and copepodites after
Effect M.A. Hashmi
Identification of Gestagen(s) and Corticosteroid(s) from Danube River water
hydrophobicity.
continuous enrichment. Specific handling tools and concentration models were applied for each fragrance tested to determine the appropriate loading conditions of static mode to prepare the test solutions for the set up a global strategy to prepare solutions of hydrophobic substances using concentrations. To increase the robustness of the cell culture medium with Oasis HLB was successfully tested and compared with the direct passive dosing using Oasis HLB. On the one hand, the medium pre-equilibration approach enables one to control the role of temperature on the equilibrium state. On the other hand, due to the fact that the dosing phase is not directly introduced in the assay, maintaining of the test concentration over the test duration is diminished for some compounds. In summary, the application of Oasis HLB as a passive dosing phase was successfully established and medium pre-equilibration for re-establishing field mixtures in an exposure medium was tested. This opens up the possibility of recreating broad mixtures sampled with Oasis HLB at natural ambient concentrations in toxicity and other tests.

TH212 Passive dosing strategy for in vitro test systems: static concentration generator and continuous release F. Begnaud, Firmenich / DRAP; C. D debonneville, Firmenich / Research and Development; V. Laubscher, F. Berthaud, Firmenich SA / DRAS; H. Schug, Eawag - Swiss federal institute of Aquatic Science and Technology / Environmental Toxicology; C. Kropl, University of Bern / Centre for Fish and Wildlife Health; K. Schirmer, Eawag / Environmental Toxicology; H. Segner, University of Bern / Centre for Fish and Wildlife Health; S. Gimeno, Firmenich / Product Safety and Regulatory Affairs
The ability to generate a true solution of a chemical substance at controlled concentrations is essential to generate meaningful aquatic toxicity information. This is especially true for in vitro test systems, and becomes highly challenging when dealing with hydrophobic (logKow > 3.5) and volatile substances like some fragrances. Historically, solvents were used to enhance the solubility, but their potential impact on results (e.g. could impact transmembrane permeation, increase bioavailability, disrupt enzymes, generate oversaturated solutions) triggered the search for alternative solutions. Passive dosing has proven to be effective to generate solutions of truly dissolved substances at controlled and constant concentrations. To increase the robustness of in vitro alternative approaches, involving permeation and biotransformation, to the fish bioconcentration test, we set up a global strategy to prepare solutions of hydrophobic substances using customized PDMS-reservoirs. These tube-shaped reservoirs were used either in static mode to prepare the test solutions for the in vitro biotransformation test with rainbow trout S9 or hepatocytes, or in dynamic mode to maintain a constant concentration in a selected compartment of the permeation setup. The strategy was applied for each fragrance tested to determine the appropriate loading conditions of the tubes to reach a defined concentration in the test media at a controlled temperature, and when necessary for the tube to act as infinite reservoir for continuous enrichment. Specific handling tools and concentration models were used to improve the throughput of the tubes preparation. We present here this strategy and corresponding results for selected fragrance compounds with varying hydrophobicity.

TH213 Identification of Gestagen(s) and Corticosteroid(s) from Danube River wastewater sample by using LC-HRMS and non-target screening approach M.A. Hashmi, M. Krauss, Helmholtz centre for environmental research - UFZ / Effect-Directed Analysis; B. Escher, Helmholtz Centre for Environmental Research GmbH - UFZ / Cell Toxicology; W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis
Biologically active substances (generally termed as endocrine disrupting chemicals (EDCs)) are present in untreated municipal wastewater, which may cause deterioration of freshwater ecosystem due to their potential to disrupt the endocrine system of aquatic organisms. Untreated municipal wastewater is directly discharged into Danube River, Novi Sad, Serbia and the objectives of this study is to identify compounds responsible for endocrine and non-endocrine effects in Danube river water by using non-target screening. Water sample from Danube River were extracted on-site using large volume solid phase extraction (LVSPE) and was pre-screened on genetically modified bioassays for agonistic and antagonistic hormonal activity for progesterone and glucocorticoid receptors (PR and GR). The extracted fractions were analyzed using liquid chromatography on a reversed phase column and the potential of causing different toxic responses towards humans and wildlife. Recent research assessment still generally rely on chemical analyses only; however, such investigations do not provide information regarding the interactions between chemicals including their integrated toxicity. The limited knowledge of the risks associated with mixture toxicity is the starting point for this study, and is part of the EnForce project that aims to investigate the toxic responses of mixtures of pollutants and integrate those results into risk assessment. In this study, we used the non-target screening approach to unravel the compounds responsible for estrogenic and corticoid activity, non-target screening is being performed by using LC-HRMS.

TH214 Mixture Risk - Development of an effect-based chemical risk assessment strategy for sites contaminated with complex mixtures of organic and inorganic contaminants G. Nilen, B. Holms, M. Larsson, Orebro University / M. Technology-Environment research centre (MTM); N. Scherbak, Orebro University / School of Science and Technology, Life Science Centre; M. Engwall, Orebro University / Man-Environment-Environment research centre (MTM); S. Keiter, Orebro University / MTM Research centre
Environmental contamination is usually comprised of a mixture of pollutants, each of them bearing the potential of causing different toxic responses towards humans and wildlife. Recent risk assessments still generally rely on chemical analyses only; however, such investigations do not provide information regarding the interactions between chemicals including their integrated toxicity. The limited knowledge of the risks associated with mixture toxicity is the starting point for this study, and is part of the EnForce project that aims to investigate the toxic responses of mixtures of pollutants and integrate those results into risk assessment. In this study, we used the non-target screening approach to unravel the compounds responsible for estrogenic and corticoid activity, non-target screening is being performed by using LC-HRMS.

TH215 Analyzing chemical pollutants in water samples from an urban river and wastewater effluent in Hyderabad (India) and their ecotoxicological effects using effect-directed analysis (EDA) J. Daniel, RWTH Aachen University; P. Böhm, RWTH Aachen University / Department of Ecotoxicology; J. Altheim, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis; A. Dayakar, Gamana R. Prasad, Harshad; A. Sambasiva, Nilaya. Inorganic Institute of Technology Madras / Department of Civil Engineering; V. Schiller, RWTH Aachen University / Department of Ecotoxicology; M. Krauss, Helmholtzcentre for environmental research - UFZ / Effect-Directed Analysis; S.E. Crawford, RWTH Aachen University / Institute for Environmental Research, Dept. of Environmental Analysis; S. Schiwi, RWTH Aachen University / Department of Ecotoxicology Analysis; T. Schulze, W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis; H. Hollert, RWTH Aachen University / Institute for Environmental Research
In India, surface water contamination in urban areas is a common issue. One major source of pollution may result from the discharge of treated and untreated wastewater, both domestic and industrial in receiving environments. This contamination composed of a complex mixture containing e.g. polychlorinated biphenyls (PCBs) and polycyclic aromatic hydrocarbons (PAHs) from industries or pharmaceuticals from residential waste may pose a risk not only to the environment but also human health. Previous studies have reported a strong presence of
multi-resistant bacteria in the Musi River, which might be due to large pharmaceutical production located in Hyderabad. A cooperation between the Department of Ecosystem Analysis RWTH Aachen (ESA), the Helmholtz Centre for Environmental Research Leipzig (UFZ) and the Civil Engineering Department from the Indian Institute of Technology Madras (IITM) was formed to evaluate the water quality in the Musi River, an urban river in Hyderabad (Telangana state, India). To avoid sustainable water management, the ecotoxicological state of the Musi River, water samples (40-100 L per sampling site) were extracted using a novel device for onsite large-volume solid phase extraction (DOI: 10.1016/j.scitotenv.2016.12.140). Two samples were taken along the Musi River, one from a tributary, one from a wastewater treatment plant effluent and another sample from an industrial effluent. Currently, these samples are screened for their toxicity using the water extracts in a set of different bioassays to select samples that are suitable for an effect-directed analysis (EDA) study. Considered endpoints are algae growth inhibition, acute daphnia immobility and mechanism-specific endpoints such as estrogenic activity and genotoxicity. In combination with target and non-target chemical analysis, the overall goal of the EDA study is to identify the main toxic drivers in one sample. Preliminary results show an adverse effect on the water flea Daphnia magna and the freshwater algae Pseudokirchneriella subcapitata. Estrogenic activity was induced in four out of five samples in the lyticase yeast estrogen screen. Further evaluation of the data and investigation on genotoxicity using the Ames assay is needed to make a well-founded decision on which assay, and sample are most suitable for EDA. Results from this work will provide insight into the composition of chemical pollutants in an Indian urban and riveric ecological effects.

TH216 Ecotoxicological assessment of water samples from an urban river, wastewater treatment plant effluent and industrial effluent in Hyderabad (India) using a set of different bioassays
P. Devi, J. Daniel, RWTH Aachen University / Department of Ecosystem Analysis; J. Althoff, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis; A. Dayakar, Gamana NGO Hyderabad; A. Sathish Lekha, J. Vijayan, I.M. Nambi, Indian Institute of Technology Madras / Department of Civil Engineering; V. Schiller, RWTH Aachen University / Department of Environmental Research UFZ / Directed Analysis; S. E. Crawford, RWTH Aachen University / Institute for Environmental Research, Dept. of Environmental Analysis; S. Schiwy, Institute of Environmental Research-RWTH Aachen / Department of Ecosystem Analysis; T. Schulze, W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis; H. Höltert, RWTH Aachen University / Institute for Environmental Research UFZ / Volume Solid Phase Extraction. Contamination of surface water is a common issue in urban areas of India. Large proportions of urban river water may consist of untreated wastewater, both domestic and industrial. The city of Hyderabad (Telangana state, India) has large industrial clusters including pharmaceutical, dye and battery factories that have the potential to affect surrounding waterbodies. Recent studies on antibiotic resistances proposed pharmaceutical industries as a potential cause for antibiotic resistances in bacteria in surface waters of Hyderabad. Daily contact of cattle and cattle herding shepherds as well as monsoon flood events are only two examples in which the river pollution is not only an environmental risk but also a human health issue. To work towards a more sustainable water management in urban areas of India, a cooperation between environmental engineers from the Indian Institute of Technology Madras and ecotoxicologists from the Department of Ecosystem Analysis (ESA), RWTH Aachen University and environmental chemists from the Helmholtz Centre for Environmental Research Leipzig (UFZ) has been formed. For a complementary assessment to previous studies, water samples were extracted from an urban river (Musi river), it’s tributary, the effluent of a wastewater treatment plant and industrial wastewater, in Hyderabad in June 2017. The samples were taken using a novel device for onsite large-volume solid phase extraction (DOI: 10.1016/j.scitotenv.2016.12.140) with a defined extracted water volume between 40 and 100 L per sample over 4 to 8 hours. The resulting water extracts will be investigated through non-target and target chemical analysis as well as effect-directed analysis (EDA). A bioassay battery to investigate the toxicological effects of the samples included algae (Pseudokirchneriella subcapitata) growth inhibition, daphnia (Daphnia magna) immobilization, fish embryo toxicity (Danio rerio), endocrine disruption (lyticase Yeast Estrogen Screen; ER-CALUX), genotoxicity (Ames fluctuation; micro nucleus test), neurotoxicity (D. rerio) and dioxin-like activity (micro EROD), these tests are currently ongoing. Preliminary results indicate adverse effects on P. subcapitata, D. magna, as well as endocrine disruption in the lyticase Yeast Estrogen Screen in four out of five samples. The combined results of this work will provide a comprehensive ecotoxicological characterization of an urban Indian river and potentially raise awareness of possibly related risks.

TH217 NASA SPEZ 2015: Ecotoxicological risks in five small Swiss streams within agricultural catchments
M. Langer, Centre Ectoxo EAWAG-EFPL / Aquatic Ecotoxicology; M. Jungbluth, Centre Ectoxo EAWAG-EFPL; S. Spycher, Eawag Swiss federal Institute of Aquatic Science and Technology; M. Koster, Amit für Umwelt, Thurgau / Gewaesserqualitaet; C. Baumgartner, AquaPlus; E. Vermeirssen, Ecotox Centre Eawag-EFPL / Aquatic Ecotoxicology; I. Werner, Ecotox Centre Eawag-EFPL / Department of Anatomy Physiology and Cell Biology
The Swiss National Monitoring of Surface Water Quality (NAWA) is occasionally complemented by focused studies on relevant topics. The latest focus study evaluated pesticides in small streams in catchments near agricultural land use. For this purpose five small streams were sampled from the beginning of March to the end of August using half-day composite samples and an analytical method covering 213 active substances. The chemical analysis was complemented with several biological investigations. Measured concentrations of pesticides were used to determine the risk of pesticide mixtures. Using acute and chronic effect-based water quality criteria (QC), we calculated risk quotients (RQ). In a second step RQmix of individual compounds were summed to provide separate mixture RQmix for plants, invertebrates and fish. In all the tested water bodies a chronic mixture risk with RQmix > 1 was determined. There was a chronic mixture risk at three sites for almost the entire sampling period. Consequently this resulted in no recovery time for aquatic organisms. An acute mixture risk was determined for four streams, with two of them showing high risks with RQmix greater than 10. In addition, pesticide effects were investigated to what extent the predicted mixture risk of herbicides was consistent with endpoints that are determined in the combined algae test. This test provides information on the inhibition of photosystem II (PS II) and algae growth. The resulting risk corresponded very well with the calculated mixture risk for PS II inhibitors. In one stream PS II inhibiting plant protection products dominated the ecotoxicological risk assessment of Lake Mondsee was carried out. Water (W) and sediment (S) samples were collected from Lake Mondsee and the reference site, Lake Irsee, on three different seasons: 1) summer 2015 (preliminary assessment of W and S samples’ toxicity), 2) spring 2016 (possible best-case scenario, since lake was frozen for the winter) and 3) summer 2016 (worst-case scenario, tourist activities peak). The WTP inflow and outflow, plus pre-thickening (PS) and a thickening (TS) sludge was also collected. The toxicity assessment for summer 2015 was made by performing 48-h population growth (r) assays with the rotifer Brachionus calyciflorus (W samples) and the 15-min luminescence inhibition assay with the bacterium Vibrio fischeri (all samples). Regarding the W samples, results showed no luminescence inhibition for V. fischeri and average r inhibition rate (%) of B. calyciflorus was below 26%. The WTP inflow samples presented high toxicity to B. calyciflorus (EC50 > 40%). Samples of S, PS and TS were extremely high toxic to V. fischeri. The samples collected during spring 2016 and summer 2016 were analysed through a battery of assays, with species belonging to different trophic levels. In addition to the two above mentioned tests: the 72-h growth inhibition test with Raphidocelis subcapitata and the feeding inhibition test was performed. Reduced survival was observed for the 6-day mortality and growth assessment with Heterocyris incongruens for S, PS and TS samples. Regarding spring 2016, the average r inhibition rate (%) of B. calyciflorus was lower than 30% for most of the W samples and average did not surpass 12%. The Microtox® tests showed high toxicity only for all W, PS and TS samples. Samples collected in the summer 2016, showed similar results for the growth inhibition for R. subcapitata. The r-inhibition rate (53%) of B. calyciflorus and the 6-day mortality and growth assessment with H. incongruens showed some variation. No evidences of the influence of the WWTP present at lake Mondsee were retrieved, since W and S samples from both Lakes Mondsee and Irsee showed similar toxicity. Further chemical analysis is necessary to clarify the high toxicity observed in the sediments.

TH219 Availability of estrogens applied onto 96-well plates in the LYES
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TH222 Bioassays stress the ecotoxicological differences between polymers and plastics additives in the marine environment

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Commercial objects made of plastics are composed of two different components with dissimilar ecotoxicological properties, namely the polymer matrix and the chemical additives used to provide the final physical and chemical properties demanded by the consumers. Most conventional polymers are made of innocuous monomers (olefins, terephthalates), they are inert under environmental conditions and, according to standard ecotoxicological bioassays using early life stages (ELS), do not pose ecotoxicological risk to marine organisms, with the possible exception of mechanical damage. In contrast, many common plasticizers (e.g. orthophthalates), flame retardants (polybrominated and organophosphorous chemicals), UV filters (benzophenones and other aromatics) and biocides (triclosan) have shown sublethal toxicity for the reproductive and endocrine systems of aquatic organisms. Those potential effects are difficult to test in laboratory since they may show long exposure times and plastic-organism interactions not considered in standard toxicity tests. Using ELS of marine organisms, we have adapted standard bioassays with ELS, tested ‘virgin’ microparticles of conventional polymers (PE, PS, PVC) and did not find any relevant short-term toxicity. In contrast, when microparticles obtained from commercial plastic objects are used events of acute toxicity are found, pointing at the additives as the causal agents of the toxicity found. Ongoing experiments explore the kinetics of additive leaching and resulting toxicity in order to assess the relevance of the results under environmental conditions. In addition, some commonly used chemical additives of plastics were also tested and some of them did show acute toxicity at levels not far above those found in polluted coastal waters. The overall experimental evidence obtained so far using standard bioassays with ELS of marine invertebrates point at certain chemical additives as ecotoxicologically unacceptable and stresses the need of finding non-toxic alternatives useful for the industry.

TH223 EFFECTS OF POTASSIUM BROMATE ON THE EMBRYOLOGICAL DEVELOPMENT OF THE SEA URCHIN Arbacia lixula (Linnaeus, 1758)

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Potassium bromate is a powerful oxidizing agent that chemically ages flour much faster than ordinary air. Potential long-term exposure times and plastic-organism interactions not considered in standard toxicity tests. Using ELS of marine organisms, we have adapted standard bioassays with ELS, tested ‘virgin’ microparticles of conventional polymers (PE, PS, PVC) and did not find any relevant short-term toxicity. In contrast, when microparticles obtained from commercial plastic objects are used events of acute toxicity are found, pointing at the additives as the causal agents of the toxicity found. Ongoing experiments explore the kinetics of additive leaching and resulting toxicity in order to assess the relevance of the results under environmental conditions. In addition, some commonly used chemical additives of plastics were also tested and some of them did show acute toxicity at levels not far above those found in polluted coastal waters. The overall experimental evidence obtained so far using standard bioassays with ELS of marine invertebrates point at certain chemical additives as ecotoxicologically unacceptable and stresses the need of finding non-toxic alternatives useful for the industry.

TH221 Determination of Izmir Bay pollution by using genetic biomarkers in the mussel Mytilus galloprovincialis

A. D. Olbrich, University Ege / Hydrobiology; H. Parlak, Ege University; M. Boyacioglu, Ege University; M.A. Karaaslan, University of Ege; G. Gilsiever, Ege University

Izmir and Manisa, has been polluted nearly 50 years. Aquatic ecosystems were affected very badly due to much kind of pollutants such as heavy metals, polycyclic aromatic hydrocarbons (PAH), Polychlorinated Biphenyls (PCB) and Pesticides. Besides agricultural and industrial activities, heavy marine transport and dredging activities in the harbor activities are also disturbed Izmir bay. Authorities have decided to take serious action when the effects of pollution were being unbearable in 1980s. As all the city smell very badly. Micronuclei (MN) tests is a system of micronuclei testing used to detect the polychlorinated hydrocarbons and drops in DNA levels. The MN formation is the first consequence occurring in the aquatic organisms. In this study, we aimed to investigate the genotoxic effects of Izmir Bay by detecting the MN frequency changes in the gills of mussels (Mytilus galloprovincialis). According to our results MN frequency of 10 stations varied between 0%, 39.33% - 5.6% and Binucleated cells frequencies were 0.17 – 5.27. Pollution of Izmir Bay is a long story and cannot be healed in short time but there are some signs that it is in the healing trend. Key Words: Izmir Bay, Pollution, micronucleus, Mytilus galloprovincialis

TH220 Mutagenic and ontogenetic responses in freshwater guppy Poecilia vivipara chronically exposed to waterborne sodium dodecyl sulfate (SDS)

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The deposition and persistence of some chemicals in aquatic environments is a constant threat to aquatic organisms. Sodium dodecyl sulfate (SDS) is a surfactant widely used as an emulsifier in household products and is constantly present in the environment (Marina et al., 2012). Studies have revealed that SDS concentration causes reproductive anomalies (micronucleated cells, nuclear buds, binucleated cells and cells with morphological analyzes. After exposure, females’ blood was analyzed for genetic changes in DNA fragments such as micronuclei in the cytoplasm of interphase cells. Damage caused on the DNA by genotoxic pollutants is the first consequence occurring in the aquatic organisms. In this study, we aimed to investigate the genotoxic effects of Izmir Bay by detecting the MN frequency changes in the gills of mussels (Mytilus galloprovincialis). According to our results MN frequency of 10 stations varied between 0%, 39.33% - 5.6% and Binucleated cells frequencies were 0.17 – 5.27. Pollution of Izmir Bay is a long story and cannot be healed in short time but there are some signs that it is in the healing trend. Key Words: Izmir Bay, Pollution, micronucleus, Mytilus galloprovincialis

TH211 Determination of Izmir Bay pollution by using genetic biomarkers in the mussel Mytilus galloprovincialis

A. D. Olbrich, University Ege / Hydrobiology; H. Parlak, Ege University; M. Boyacioglu, Ege University; M.A. Karaaslan, University of Ege; G. Gilsiever, Ege University

Izmir Bay, which is surrounded by many agricultural and industrialized cities like
Effect of thermal stress on endocrine disruption in Daphnia magna
J. Na, Korea University; H. IM, J. Jung, Korea University / Environmental Science and Ecological Engineering

Endocrine disrupting chemicals (EDCs) include various types of natural (17β-estradiol, estrone) and synthetic (nonylphenol, bisphenol-A) compounds presenting inhibition or mimicking of the reproductive action of endocrine system in men and humans. Recently, several research studies indicated that daphnids are species which reproduce by parthenogenesis and may generate offspring in response to EDCs. In addition, it was demonstrated that variation of water temperature is able to change reproduction, growth, and survival of aquatic organisms and population number.

This study aims to evaluate the effect of thermal stress on endocrine disrupting effect of EDCs using Daphnia magna. Short-term screening (STS) assay was used to compare the endocrine disruption effects using adult (10–17 days old) daphnids. Animals were exposed to two temperatures of 20 °C and 25 °C, and reproduction, growth, male production and survival rates were evaluated. This study can give an insight into the endocrine disruptive effects of EDCs on aquatic organisms under influence of thermal effluents discharged into streams and rivers.

TH225

Microplate Algae Growth-Inhibition Bioassay
I. Iturria, O. Jaka, C. Martí, A. Alzualde, BioBide; A. Muriana, BBD BioPhenix S.L. / RD

The development of new chemical compounds is a long and costly project that may span up to 10 years. However, the success rate of new chemical families has decreased exponentially in the last decades mainly due to compounds toxicity detected in later phases of the R&D process. The OECD publishes a series of guidelines to define the toxicity evaluation assays required for regulatory purposes. The 201 guidelines, in particular, describes the alga growth inhibition test for the evaluation of aquatic toxicity. Nevertheless, the procedure is tedious and time-consuming, so it’s not suited for high throughput screening of toxicity on early development phase. Given so, there is a need for new fast and cost-effective assays with an increased throughput to assess the aquatic toxicity of a compound in early phases of the development. In this work, we present a miniaturized version of the OECD 201 alga growth inhibition test. The miniaturized test is carried out in 96 well plates and the biomass measurement is performed on a plate reader. The methodology makes possible to test ten concentrations of a compound and a negative control on the same plate. The biomass measurement by fluorescence read produces a sensitive and reproducible measurement of alga concentration in an efficient manner, with a significant time and human labor reduction. The alga microplate assay was validated with environmentally relevant reference compounds (such as pesticides or flame retardants) and the resulting IC50 values were compared to the OECD 201 results.

TH228

Sustainable Guar Initiative - an integrated approach of social and environmental LCA
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Sustainable Guar Initiative (SGI) is a three-year long integrated program aiming at developing sustainable guar production within the Bikaner district in Rajasthan, India. This desert district is one of the largest producers of guar and guar gum in India. SGI was set up by Solvay, L’Oréal, HiChem and the NGO TechnoServe, and is based on 4 themes: (1) Agronomy: enhancing sustainable practices for rain-fed guar production, (2) Environment: groundwater-neutral approaches and best practices in guar farming, along with tree plantation, (3) Social impact: gender approaches, nutrition, health & hygiene and (4) Market improvement: traceability, supply chain and market access. Guar gum is extracted from guar seed and can be used as such, or functionalized. It is for example used as a bio-based thickening agent in personal care products. To confirm and consolidate the relevance of the program and to identify potential improvement opportunities, an environmental and social Life Cycle Assessment (LCA) has been conducted, comparing the guar production before and after the SGI. The social LCA has been conducted following already available guidelines, including UNEP-SETAC Guidelines for Social Life Cycle Assessment of Products and WBCSD Social Life Cycle Metrics for Chemical Products. Methodological developments have furthermore been undertaken in order to fully take into account the smallholders. The Master’s thesis of Diana Indrane on “Integrating Smallholders within Handbook for Product Social Impact Assessments” has been completed with some developments, related to: (1) Goal and scope: better identification of relevant stakeholders and social aspects, (2) Inventory: improvement of data quality among the social aspects, stakeholders or life cycles steps and (3) Performance assessment: common rating system enabling aggregation related to inventory from multiple sources. Besides environmental LCA, the poster will focus on social LCA. Methodological challenges encountered will be presented and solutions to tackle them will be detailed as long with other limits related to these new developments.
(modelled technoeconomically as part the Horizon 2020 project REFLEX). It has been developed in light of previously published work aiming at life-cycle based social and/or environmental assessment of single energy technologies and energy systems with a future perspective, and in careful collaboration with the handful of REFLEX partners responsible for energy systems models. The functional unit for the assessment of the future scenarios is the provision of energy services in the EU in 2050 (the final year of the scenario and modelling). The system boundary for the energy systems models proposed that the entire energy system is encompassed, including all energy flows and associated supply chains required for the production and delivery of heat (in all sectors), electricity and mobility services. Foreground LCI data for the capital equipment (primarily plant and vehicles) and fuels are taken from output data from energy systems models. Background LCI data about separate future energy technologies (as requested by the identified foreground processes noted earlier) start from existing LCI data for current systems (from Ecoinvent) to which changes are made based on certain assumptions about future developments of the technology in question. Thanks to the availability of price data for material in the latest version of Ecoinvent, LCI data acquired can be converted into monetary values. Finally, the cost data can be inputted into a multi-region IO-table linked worker hours model such as social hotspots database. This yields the country specific sectors required for the material in the energy system. The social impacts are then be evaluated with social theme tables for each country specific sector. It is proposed that IO-tables used should be adapted in order to reflect the development of the energy system in the future. Since the energy models and scenarios used in REFLEX are preserving in nature, it is suggested as a simplification that social impacts for the future system models are predefined for the countries and the stage of operation that they perform, without any further adaptation that is necessary for such results should and should not be interpreted. The methodology will be operationalized in the coming year as part of the REFLEX project.

TH231 Social Life Cycle Assessment of the water system in Mexico City M. Garcia, Instituto de Ingenieria, UNAM / Ingenieria Ambiental; L. Guereca, Engineering Institute Universidad Nacional Autonoma de Mexico / Environmental Engineering One of the main elements of the sustainability of water systems in the cities, is to guarantee a decent job that promotes the welfare of workers in accordance with the objectives of sustainable development in Agenda 2030. Mexico City is one of the most populated cities in the world and is considered as the main political, economic and cultural centre of Mexico. However, it has been several water sustainability problems in the social aspect as risks to the health of workers of the water system. The activities of operation that they perform, are also subject to the social system of the water system such as professional development, and aging of the labour force. The objective of this research was to carry out an assessment of damages to human welfare of the workers, through a holistic and systemic approach to assess the impacts of each of the processes of the water system in relation to working conditions. The evaluation of the social impacts of the water system was based on methodological guidelines of S-LCA edited by UNEP/SETAC/LCI and other instruments of social impact assessment. However, if considering five stages of the water system: water abstraction and treatment, distribution, storage, waste water collection and wastewater treatment. The evaluation used the method of impact assessment with a nominal scale between 0 and 1, divided into five ranges of social performance: Without information, Bad, Medium, Good and Very Good. The results were that Water abstraction was the stage with the highest performance, good water quality and good governance Distribution. The Storage was the stage with the lowest value of social performance with a level of Medium. While, in the stages of the wastewater management, the stage of Waste water collection obtained a performance of Medium and the stage of Wastewater Treatment, with a performance of Good. Any stage of the system has reached a Very good level in social performance. In conclusion according to the methodology used, which adopts a scale of 0 to 1, where 0 is the worst and 1 the best; it determines a score of 0.6 for all the analysed system, which places it in a Medium position. However, the most critical stage was Storage with a score of 0.4, which suggests that improvements are needed in this stage to achieve a better social performance.

TH232 Environmental Risk Assessment for some additives used in hydrocarbon extraction activities into the sea S. Santoro, National Research Council of Italy (CNR); S. Giardina, Ministry for the Environment, Land and Sea; M. Orrù, National Center for Chemical Substances - National Institute of Health; D. Romoli, Italian National Institute for Environmental Protection and Research. Concerning the oil and gas offshore platform activities, the Italian Ministry of the Environment, Land and Sea has adopted a new approach to decide for the release/renewal of the authorisation to discharge the Produced Formation Water (PFW), a by-product of both oil and gas extraction, into the sea. This approach aims at assessing more deeply the possible environmental impact of the additives used in hydrocarbon extraction activities. In this context, we present the application of the environmental risk assessment methodology, set out by REACH Regulation on chemicals, for some additives (e.g. Diethylene glycol) used in oil and gas platform activities. This approach allows us to determine specific concentration levels eligible for seaward discharge of these additives. By applying this methodology we compared the Predicted No-Effect Concentration (PNEC) with the predicted environmental concentration (PEC) related to the release of the substance in the seawater. The work already concluded on Diethylene glycol showed that the concentration considered, having been determined at 670 µg/l and 680 µg/l for constant/frequent release and 5900 µg/l for intermittent release. These limits have been included by the Ministry of Environment as a binding condition for granting the authorisation of discharge to the platform manager. Session: 3.12 Improvements in environmental exposure assessment: development and application of tools industry sectors, regulatory agencies and international boundaries. Authors: Serafina Santoro – National Research Council (CNR) - Institute of Atmospheric Pollution Research Italian Ministry of the Environment, Land and Sea Silvia Giardina – Italian Ministry of the Environment, Land and Sea – General Directorate for environmental assessments and authorisations Maria Antonietta Orrù – National Institute of Health - National Center for Chemical Substances Debora Romoli - Italian National Institute for Environmental Protection and Research.

TH233 Multidisciplinary approach for discussing the rice crop specific needs in Southern Europe in the view of the Plant Protection Products assessment: conclusions from an ad hoc workshop A. Carone, Dow AgroSciences Italia srl; A. Whyte, Dow AgroSciences Ltd; A. Aix, Dow AgroSciences / Risk Management; G. Azimonti, ICPS International Centre for Pesticides and Health Risk Prevention; R. Cabella, INAIL - Dipartimento di Medicina, Epidemiologia, Igiene del Lavoro e Ambiente; G. Canha, Lussoem A.S.A.; C. Civitiella, T. Corinti, ICPS International Centre for Pesticides and Health Risk Prevention; M. Corvaro, Dow AgroSciences Ltd; N. Dalla Valle, Dow AgroSciences Italia srl; F. Dias, Cooperativa Agrícola Montemor; E. Garcia, I. Gonzalez, Dow AgroSciences Ibérica, S.A.; M. Guarise, Dow AgroSciences Italia srl; P. Havens, Dow AgroSciences LLC; M. Luini, F. Marchetto, ICPS International Centre for Pesticides and Health Risk Prevention; G. Mereggalli, Dow AgroSciences Italia s.r.l / Ecotoxicology; M. Osuna Ruiz, CAS, C. A. Ritter, Waterborne Environmental; M. Santoro, National Research Council (CNR) - Istituto Superiore di Sanità; C. Thomas, Centre Francais du Riz; I.S. Travlos, Agricultural university of Athens; S. Ullucchi, ICPS; W. Williams, Waterborne Environmental, Inc.; V. Zaffagnini, C. Vaj, Dow AgroSciences Italia s.r.l. In Europe and in the context of Regulation (EC) 1107/2009 for placing into the market of plant protection products rice as a crop is an anomaly and has created several difficulties in its evaluation. For regulators, there is a need for comprehension of the unique agronomic practices, application techniques, water management and environmental concerns, both from an ecotoxicology and environmental fate perspective considering the majority of rice cultivated within the Europe is grown in paddy fields. This gap in knowledge caused issues in the evaluation of rice as a representative use in the process of European authorization of active substances for plant protection products and raised questions over the suitability of environmental indicator species and risk assessments within the context necessary. Rice is a major crop in many Southern Zone Countries and the difficulties gaining an understanding of rice practices, compounded by uncertainty with changing regulatory requirements and a lack of transparency in evaluation procedures has hampered the process of active substances approval. Such a complex framework could dissuade active substance renewal by agrochemical manufacturers or indeed inhibit innovation. In conjunction, an increase in weed resistance to plant protection products currently available has forced Member States to continually rely on Emergency Uses Permit year on year to support the rice growing community. Weed and pest tolerance to agrochemicals is increasing across Europe. In an attempt to avoid problems such as the lack of adequate tools available for weed and pest control, Rice farmers recognise the urgent need for active substances to be placed on the market with different modes of action to combat resistance and safeguard the production. The above mentioned topics have been deeply discussed among experts of different disciplines from the rice producing European Countries in an ad hoc workshop facilitated by Dow AgroSciences in July 2017. The outcome of the discussion highlighted the benefits of a multidisciplinary, holistic approach, with farmers, local networks, users and research institutes facilitating an environment to coordinate a strategy for implementation, with a Member State authority championing this venture through zonal steering groups. The main conclusions of the workshop will be presented and discussed in the poster.

TH234 The Water Column Monitoring Program in Norway: when regulation and science meet D. Pampanin, International Research Institute of Stavanger; S.J. Brooks, NIVA
Norwegian Institute for Water Research; B. Grovik, Institute of Marine Research; E. Lyng, International Research Institute of Stavanger; R.C. Sundt, Statoil

Oil and gas companies operating on the Norwegian Continental Shelf (NCS) are required to carry out environmental monitoring to obtain information on the actual and potential environmental impacts of their activities and to give authorities a better basis for regulation. Scientists, operators and regulators have worked jointly for the last two decades in this program, for developing, communicating and implementing knowledge, methods and tools to manage the offshore produced water discharges. A multidisciplinary approach (e.g. chemistry, biology, modelling and risk assessment) has been developed to monitor the discharge and reduce risk. Potential impacts from no-targeted chemicals have also been in the aim of this extensive monitoring program for anticipating negative effects at the ecosystem level. In 2017, a new protocol was developed as result of the coordinated and concerted performed since 1995. The new requirements have been applied for the first time in the 2017 Water Column Monitoring program. This holistic approach shows a significant improvement in the scientific outcomes of the monitoring, in a cost-efficient way. The surveys included the use of species from various trophic levels and the analysis of both chemical and biological parameters. Three regions were selected: Tampen, Southern North Sea and Egersundbanken (reference area) and in addition the near platform effect (Stafljord A) was assessed. The study design included the use of a predictive discharge model (Dose-related Risk and Effect Assessment Model, DREAM). This model calculates the fate of the discharge in 4 dimensions (including time) to predict environmental concentrations, risk and effects. Biological and chemical data confirmed the accuracy of the study design and predicted risks. These information can be used to be used in the context of chemical stressors from natural variability and from impairment due to other stressors, such as biological and physical stressors. Costly remediation and litigation decisions often hinge on an assumption of causality. It is therefore essential that ERAs objectively examine all plausible causes of observed impairment and attempt to establish cause-and-effect relationships between stressors and responses. The FCSAP causality assessment framework is based on U.S. Environmental Protection Agency’s CAEDDS guidance, though it is simplified in an easy-to-use framework. The FCSAP causality assessment framework is based on U.S. Environmental Protection Agency’s CAEDDS guidance, though it is simplified in an easy-to-use framework.
TH241
A Bayesian approach to estimate biodynamic model parameters: bioaccumulation of PCB 153 by the freshwater crustacean Gammarus fossarum.

A. Ratier, Iristea Lyon; C. Lopes, Université Lyon 1, UMR CNRS 5558 / Laboratory of Biometry and Evolutionary Biology; H. Budzinski, University of Bordeaux; P. Labadie, UMR CNRS EPOC Universite Bordeaux / UMR 5805 EPOP; L. Peve, UMR CNRS EPOC LPTC; N. Delorme, H. QUEAU, Iristea Lyon / UR MALY Laboratoire Ecotoxicologie; O. Geffard, Iristea / UR MALY Laboratoire Ecotoxicologie; M.P. Babut, Iristea / Water

Toxicokinetic models are used to describe how organisms bioaccumulate chemicals or other substances according to uptake and elimination processes. They provide a theoretical framework for understanding phenomena, testing hypotheses, and predicting some outputs of interest. In these models, the absorption process can result from dissolved or trophic routes. The elimination process includes excretion, biotransformation and dilution by growth. To date, models exist to describe the accumulation of persistent organic pollutants (in aquatic and terrestrial systems) in various aquatic organisms. However, taking into account biotransformation remains problematic despite its potential importance. It is a key process that can limit the bioaccumulation of parent compounds while generating potentially hazardous metabolites. It varies considerably among species and contaminants. The aim of our study is to propose a Bayesian framework to estimate the parameters of a biodynamic model taking into account biotransformation, by considering simultaneously accumulation and depuration data. We illustrate our approach with the freshwater benthic invertebrate Gammarus fossarum exposed for 7 days to a sediment spiked with PCB153 and transferred to a clean media for 7 more days. The PCB153 concentrations in Gammarus fossarum increased from an initial concentration of 0.32 to 12.36 ng l⁻¹ (ww) at the end of the accumulation step. When gammarids were transferred into a clean media, the PCB153 concentration in organisms decreased to 6.41 ng l⁻¹ at day 14. The bioaccumulation model assuming first-order kinetics was fitted to the data using Bayesian inference. The inference process quickly converged and thin posterior distributions were obtained for each parameter, meaning that data brought enough information to estimate precisely each parameter. The median model predictions and their 95% credibility intervals showed a good fit of the model to the data.

TH242
Bioaccumulation and biotransformation of Hexabromocyclododecane (HBCD) by the freshwater crustacean Gammarus fossarum: a Bayesian approach to estimate biodynamic model parameters.

A. Ratier, Iristea Lyon; C. Lopes, Université Lyon 1, UMR CNRS 5558 / Laboratory of Biometry and Evolutionary Biology; H. Budzinski, University of Bordeaux; P. Labadie, UMR CNRS EPOC Universite Bordeaux / UMR 5805 EPOP; L. Peve, UMR CNRS EPOC LPTC; N. Delorme, L. Garnero, Iristea Lyon / UR MALY Laboratoire Ecotoxicologie; O. Geffard, Iristea / UR MALY Laboratoire Ecotoxicologie; M.P. Babut, Iristea / Water

The multimedia model Simple Treat is applied in the chemical legislation in Europe for decades and recently also in other legislations around the globe. It evaluates the distribution and elimination of chemicals by sewage treatment plants (STPs) in a municipal environment. It has been shown that the specific settings of industrial STPs differ largely compared to the standard settings of the municipal model of Simple Treat in terms of inter alia biological oxygen demand, hydraulic retention time, and nutrient removal. The Simple Treat models the chemical fate in industrial STPs (iTreat, Straijs et al. 2016, Chemosphere 159, 619-627) but failed to show site specific validation due to lack of appropriate data. Therefore, the iTreat model was parametrized to the specific parameters of an industrial STP at a specific site. Time series of measured elimination of 22 substances in the waste water stream of this specific STP were gathered and compared to the calculated elimination rates. The bioaccumulation rate constants of the substances were also compared to the non-parametrized model of iTreat and to the municipal models of Simple Treat 4.0 and Simple Treat 3.1. Overall, the iTreat model was successfully adapted to model chemical fate and behavior in an industrial, site specific STP. The elimination rates of the parametrized iTreat model were generally in better agreement to the measured elimination rates than for all other models investigated. The bioaccumulation rate constant of substances turned out as a more sensitive parameter when predicting the elimination rate with either model. In detail, substances with low biodegradation rate constants (e.g. the rate constant of 0.1 h⁻¹) in the dataset show far more realistic elimination rates in the parametrized iTreat model than for the non-parametrized iTreat or the two municipal models. Compared to the municipal model of Simple Treat 3.1, the parametrized iTreat showed about two-fold higher elimination rates which reflected the measured elimination. The application of site specific degradation rate constants (derived from biodegradation tests with adapted activated sludge of the respective site) for the site-specific model of iTreat will also be discussed in this poster contribution. Taken together, the validation exercise was successful and the parametrized iTreat model is applicable to other substances being produced at this site where measured data is not available.
Our findings show that race/ethnicity, gender, and socioeconomic status can be statistically significant predictors of chemical exposure. More specifically, parabens, which are chemicals used in personal care products (PCPs), and 2,4- and 2,5-Dichlorophenol, which can be a products of photo-degradation of tricosan, a common antibacterial and antifungal agent, were observed to elevated in African Americans when compared to White Americans. In addition, higher levels of pollutants were observed in areas with intensive agronomic activities, while men had showed higher concentrations of N,N-Diethyl-meta-toluamide (DEET). This could possibly be from women using cosmetic PCPs more frequent and in larger amounts, and men using insect repellent slightly more frequently than women do. Finally, individuals of higher socioeconomic status had higher levels of benzophenone-3 (used in sunscreen products), parabens, and tricosan, which could possibly be explained by more accessible variability to PCPs. In this study, we have identified inherent and demographic traits associated with elevated biomarker concentrations. We hypothesize that this is due to use patterns of consumer product, particularly PCPs. This could support research findings emphasizing the importance of near-field chemical exposures.

**TH244**

**Occupational exposure to flame retardants among Canadian e-waste dismantlers**

L.V. Nguyen, University of Toronto - Scarborough / Department of Physical and Environmental Sciences; V.H. Arrandale, Cancer Care Ontario; M.L. Diamond, University of Toronto / Department of Earth Sciences

The amount of e-waste produced globally is growing dramatically. National numbers suggest that in 2018, 20.4 million metric tons of e-waste were generated, and of these, 60% is generated in developed countries. In developed countries, e-waste is predominantly a heterogeneous mix of hazardous and non-hazardous waste, which makes it difficult to manage. The amount of e-waste in developed countries is due to use patterns of consumer products, particularly personal care products (PCPs), and the concomitant pollution with absorbance detection is far below than needed. In Italy quality vines are sometimes grown in small fields on steep slopes where spray-gun application of pesticides is used, a technique that is very costly and labor intensive. A possible alternative is the use of a fixed spraying system, and first researches are in progress. A fixed spraying system prototype was built in a vineyard in Lambrigh Research Centre and a trial was performed with the aim of measuring the deposition pattern of droplets on the roof and between rows with water sensitive papers, also in comparison with a precise low-drift air-blast sprayer. Results show that a fixed spraying system has the potential to apply plant protection products without generating drift problems, with a performance similar to a low-drift sprayer, becoming an opportunity for vineyards on very steep slopes.

**TH245**

**Global approaches to environmental exposure - assessment of e-wastes**

D. Purchase, Middlessex University / Department of Natural Sciences, Faulty of Science and Technology; L. Bisschop, Erasmus University Rotterdam / Department of Criminology; C. Ekberg, Chalmers University of Technology / Division of Energy and Materials, Department of Chemical and Chemical Engineering; P. Fedotov, Russian Academy of Sciences / Vernadski Institute; H. Garchik, Middlessex University; N. Kandile, Ain Shams University / Department of Chemistry, Faculty of Women; R. Luque, University of Córdoba / Chemistry; O. Popoola, Yaba College of Technology / Department of Chemical Science; H. Ruedel, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Department of Environmental Monitoring; A. Serpe, University of Cagliari / Department of Chemistry and Architecture; K. Surati, Sardar Patel University / Department of Chemistry; B.P. Wilson, Aalto University / Department of Chemical and Metallurgical Engineering.

Obsolete or end-of-life electrical and electronic equipment waste streams continue to grow exponentially, creating a worldwide pollution problem. E-waste comprises a heterogeneous mix of hazardous and non-hazardous metals, metalloids, glass, plastic, flame-retardants, flame-retardants, rare earth metals, metalloids, and non-metallic materials. In developed countries, e-waste is managed by using two major strategies: either by internal recycling/disposal or via exportation to developing nations. For developing countries, the management of e-waste is complicated by illegal waste shipments and further exacerbated by weak environmental regulations coupled to inadequate technology and organizational structures. Rudimentary methods such as dismantling, chipping, melting, and burning are often used by the informal sector to recover valuable materials from different e-waste components. These unofficial recycling practices contribute to the release of toxic metals and persistent pollutants that affect both the environment and human health. As a result, e-waste issues are complex, multi-faceted and can only be successfully tackled via a multidisciplinary, trans-boundary approach that involves all stakeholders that include amongst others: manufacturers, scientists, environmentalists, policy makers, waste professionals and consumers. The e-waste project “[The Environmental and Health Challenges of E-waste and its Management: an Emerging 21st Century Global Concern” (#2014-031-3-400)], supported by the International Union of Pure and Applied Chemistry (IUPAC), brings together multidisciplinary global experts to explore different aspects of the waste challenge: chemical analysis of contaminants, policy and governance, environmental and health impacts, development and advances in treatment technologies including e-waste valorisation. This presentation makes use of studies from the world to highlight the following: i) discrepancies in the provision and enforcement of regulations between developed and emerging countries; ii) complexity in the analysis of e-waste contaminants in environmental and biological samples; and iii) lack of harmonisation of tools or indices to assess risk in environment and health, particularly in soil. It is proposed that a harmonised approach should be taken to use appropriate specification analysis (e.g. to assess bioavailable or bioaccessible fractions) to evaluate e-waste contaminant risk.
µg/L. Here an existing continuous flow analysis (CFA) method was selected using a system with a special cuvette installation allowing a higher sensitivity. The protocol was validated and accredited according to standard ISO/IEC 17025. With this system an LOQ of 0.15 µg/L can be reached under optimal conditions while an LOQ of about 0.3 µg/L is achieved during routine operation. Previous to field testing it was verified that samples can be stabilized for at least 24 h by adjusting the pH of samples between 12 and storing it in the dark at 4 °C. Samples spiked with low concentrations of a cyanide standard were used as positive controls. The field validation results were satisfactory, confirming that the protocol is fit for purpose. Finally, samples from several sites of a small stream with low anthropogenic influences (River Lenne) were taken and analyzed. Free cyanide concentrations of up to 0.4 µg/L were detected. There were significant differences in free cyanide concentrations between sampling sites, with levels varying from below the limit of detection (LOD, 1/3 of the LOQ), and downstream sampling points where cyanide concentrations were at least 50% higher, possibly due to degraded plant biomass in the water. This first measurements revealed that background concentrations of free cyanide in the tested surface waters can be below the proposed EQS of 0.5 µg/L.

However, the analysis of further parameters (e.g. geographical regions, seasonality) is necessary to create a reliable database on the range of free cyanide background levels as basis for EQS implementation.

TH249 Application of equilibrium and kinetic passive sampling method to quantify integrative chemical profile in a small river and the outflow of WWT Y.-H. Lee, H. Kwon, H. Jeon, KIS, Taegu; S. Yoon, H. Kim, ETRI; E. Meyer, E. Fünfrocken, H. Beck, Saarland University; K. Smith, RWTH Aachen University / Institute for Environmental Research; A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics

Monitoring water quality is challenging as most of organic contaminants present at trace levels and chemical profile is fluctuating. Current legislative requirement of water quality monitoring, with levels including 8 priority substances in EU WFD, is a task of monitoring for priority substances, which is efficient approach in water management. However, the capturing variable contaminant level is also critical for eco-toxicalogical risk assessment. The present study reports on how to exploit equilibrium and kinetic passive sampler in parallel and evaluate dynamic chemical profile in order to satisfy the growing requirements and environmental risk assessment. Furthermore, partitioning kinetics of a range of organic contaminants toward each passive sampler type were discussed. The equilibrium sampler tends to be biased toward present chemical profile, which is rather close to instant sample, whereas kinetic sampler provides a time-weighted average concentration (C_TWA) over the full sampling period. PDMS sheets with two different thicknesses (76 and 205 µm), as an equilibrium passive sampler, were deployed without the application of performance reference compounds. From the concentration ratio from two PDMS sheets, true in situ concentration (C_{in situ}) of a range of target compounds was determined. In parallel, two types of polar organic chemical integrative sampler (POCIS) were selected as a kinetic passive sampler. The one is typical POCIS with Oasis HLB® sandwiched between polyethersulfone membranes and the other is modified POCIS (POCIS) is the most used and investigated device to date. Unlike POCIS, the presence of a diffusive gel may reduce the influence of the water boundary layer, and then hydrodynamic effects on sampling rates. Our objective is to develop the o-DGT for a reliable sampling of a wide range of 60 pesticides, 20 hormones and 38 pharmaceuticals in water selected to cover a wide range of physico-chemical properties (hydrophobicity, ionisability, size, functional groups, ...). For that purpose, we first chose the best diffusive gel (e.g. agarose or polyethersulfone and nylon) by determining diffusion coefficients for all the compounds, with the comparison of 2 methods: slice stacking and diffusion cell. The slice stacking consists in contaminant diffusion from 1 spiked gel to 5 clean gels let in contact for 30 minutes. The concentration in each gel disk was determined over the time, allowing the calculation of diffusion coefficients according to Fick’s second law. Diffusion coefficients obtained with this method are congruent with those found in literature. Those obtained with the slice stacking except for ionic compounds, which also exhibited lower affinity with gels than water, in comparison to neutral compounds. The second step consisted in membrane selection, necessary to protect diffusive gel, and that needs to exhibit the lowest possible compounds retention. Three types of membrane (cellulose, polyethersulfone and nylon) were tested with different pore sizes (0.45 and 0.20 mm). Finally, to estimate the sampling rates, and to validate diffusion coefficients, a calibration experiment with micropollutants at environmental concentrations was conducted.

TH251 Evaluation of Translocation of [14C]Radiolabeled Plant Protection Product in Tomato Fully Grown in a Greenhouse S. Freedlander, Smithers Viscent, LLC / Environmental Fate and Metabolism; S. RAO, Gowan Company / Regulatory; K. Malekani, S. Kang, Smithers Viscent / Environmental Fate and Metabolism

Once applied to a plant, pesticide residues have the potential to move to other plant tissues (via a phloem and xylem), to root tissue, and eventually to human consumers of the plant tissues. The objective of this study was to evaluate the translocation of a pesticide through phloem and xylem to various tomato tissues (flower, leaf, stem, and root) when applied to leaves and soil. A suspension concentration (SC) formulation was prepared with 14C radiolabeled active ingredient. The study was conducted with three groups of tomato plants. Group 1 treated with a foliar spray of 1 µg active ingredient/g leaf at petiole base, Group 2 treated with formulation containing 1¹⁴C active ingredient by foliar application and soil drench, respectively, in a single application at a rate of 0.50 kg a.i./ha. Leaves, stems, and flowers were harvested at 1, 2, 4, 6, and 8 weeks after application (WAP) and root tissue was harvested at 8 WAP. At each sampling interval, the isolated leaves, stems, and flowers were analyzed for total radioactive residue (TRR) by combustion analysis and autoradiography by phosphor-imager analysis. The TRR in all tissue types from the soil drench group was higher than in corresponding tissues from the foliar application group. The autoradiographs of all tissue types from the soil drench group were comparatively darker than in corresponding tissues from the foliar
application group. Although both basalpetal movement (downward from leaf application site) via phloem and acropetal movement (upward from both leaf and root application sites) via xylem were observed, results indicate the movement of radioactive residues is much faster through xylem. Select tissue samples were extracted and analyzed by HPLC-RAM, which shows that the majority of translocated radioactive residues by phloem was metabolites of the active ingredients. In order to determine movement of translocation during a conventional plant metabolism study can provide valuable information to better assess the potential effects of plant protection products on pollinating insects.

**TH253**

An *Examination of Microbial Biomass in Sediments and the Impact of Seasonal Variation* by

K. Malekani, Smithers Viscent / Environmental Fate and Metabolism; S.P. McLaughlin, Smithers Viscent / Department of Environmental Fate; K. Campbell, Smithers Viscent / Environmental Fate Metabolism

Microbial biomass is an important measure of the health and viability of a sediment just as it is for soils. It is also a parameter used to assess viability of the seetoxicological studies. N. Lembcke, Institute of Industrial Organic Chemistry in Chemnitz, Germany; Branch in Piszczyna, P. Ploszczyca, P. Wójcik, Institute of Industrial Organic Chemistry Branch in Piszczyna / Department of Toxicological Studies; Z. Hayez, Institute of Industrial Organic Chemistry Branch in Piszczyna / Department of Toxicological Studies

The current presentation will focus on the initial microbial biomass as an indication of health and viability of sediments at the time of their collection. A collection of initial sediment microbial biomass values has been summarized and presented based on their time of collection during the year. An example from a shallow-water sediment (Taunton) and one sandy sediment (Weweantic) collected during all four seasons, over a two-year period produced microbial biomass values (expressed as % OC) shown below. 2016 Taunton Weweantic 2017 Taunton Weweantic Winter 0.47 0.11 Winter 0.81 0.05 Spring 0.52 1.1 Spring 0.76 0.82 Summer 0.63 0.28 Summer 0.51 0.41 Fall 0.40 0.22 Fall 0.60 0.71 late summer Additional biomass results will be presented, discussed and correlated to other sediment parameters, including texture, pH, and % OC. Conclusions from several sediments used in recent years will be extrapolated from trends in the data set concerning seasonality, environmental conditions and sediment characteristics.

**TH254**

Use of scanning electron microscope (SEM) in evaluation of hypopharyngeal glands development in Honey bees (Apis mellifera L.) by


The hypopharyngeal glands (HPG) of Honey bees consist of many acini connected with a collecting duct, arranged in the form of long paired cords lying on both sides of the head. They played important role in maintaining healthy colonies i.e. through production of “milk” containing proteinaceous substances to feed larvae and queen. The aim of this study was to check the possibility of using scanning electron microscope (SEM) to evaluate the development of hypopharyngeal glands of bees, considering reliability, work-, time-consuming and cost-effectiveness of the method, including collecting of material. The study was conducted on Honey bees (Apis mellifera L.) subjected to chronic toxicity studies performed according to the EFSA guidelines (EFSA Journal 2013;11(7):3295). Animals were treated with different chemicals in 4 to 5 concentrations. The left HPG were obtained from 5 bees per test item (in the highest concentration, which did not cause mortality below 10%). After dying the bees, the HPG were taken from each samples. The analysis of the results showed decreases and increases of acini and their number per 1 mm², depending on the test item, however, these differences were not always statistically significant. It turns-out, that images were very valuable, allowing visual comparison of acini. Data obtained from the studies indicate that SEM can be useful tool for evaluation of hypopharyngeal glands development of Honey bees.

**TH255**

Comparison of International Quality Assurance and Quality Control Standards for High Resolution Mass Spectrometry Dioxin Analysis by

D. Thal, E. Ogburn, Environmental Standards Inc; R. Vitale, Environmental Standards; D. Bly, Environmental Standards, Inc.

The analysis of polychlorinated dibeno-p-dioxins and polychlorinated dibenzofurans (PCDD/Fs) in environmental media, foods and tissues by high resolution gas chromatography-high resolution mass spectrometry (HRMS) is frequently used as the reference methodology against which other candidate analytical approaches are evaluated. Official methods such as the intercalibration technology, equipment and methodologies typically labeled standard for recovery correction have been established in the EU, the USA, Japan and other nations for decades and international standards for such methods have been established by ISO (Standards 13914 and 18073, for example). To identify achievable best practices and to understand differences in precision, accuracy and qualitative certainty for data produced from wide-ranging sources, an examination of the requirements of these methods was conducted. A review identifying critical differences and areas of agreement with regard to qualitative criteria, precision and accuracy will be presented, with perspectives on the impact differences may have on data uses by researchers conducting analyses under different protocols.

**TH256**

New Mass Spectrometry Techniques for the Measurement of Persistent Organic Pollutants.

P.D. Jones, University of Saskatchewan / School of Environment and Sustainability; J. Giesy, University of Saskatchewan / Department of Veterinary and Biomedical Sciences and Toxicology Centre

Recent development of new forensic advanced technology and instrumentation has increased the amount and quality of analytical information that can be obtained from samples. In particular, dramatic increases in mass resolution have made possible unequivocal identification of contaminants even in complex mixtures and matrices. In the area of POPs analyses of PCDD/Fs and PCBs are of concern due to small concentrations that need to be quantified and the presence of a wide range of isomers that can potentially interfere. Implementation of a GC/Orbitrap system brings levels of mass resolution not previously available for analysis of POPs by GC chromatography coupled with ultra-high mass resolution mass spectrometry (GC-QUHRS). Here we report use of GC-QUHRS for identification and quantification of PCDD/Fs and PCBs. The methods developed are based on standard US-EPA methods (Methods 1613 and 1680) but are enhanced by use of new capabilities provided by image current detection and high mass resolution (>100,000 FWHM). Robustness of the PCDD/F analyses were demonstrated by excellent calibration characteristics and ability to detect all 2,3,7,8-substituted congeners even in an extract of used motor oil. Reanalysis of fish tissues previously analyzed by a magnetic sector instrument demonstrate accurate of identifications and quantifications and provide validation of the method. Analyses were therefore conducted to determine the potential for a ‘multiplex’ analysis of various POPs where the resolving power (>120,000 FWHM) of the MS system was able to eliminate potential interferences from a variety of ‘non-target’ organochlorines. These multiplexed analyses significantly reduce the time and cost of sample extract preparation and clean-up. In addition, the relatively simple auto-tuning and mass calibration algorithms available for the advanced mass spectrometry systems greatly simplify these otherwise challenging analyses.

**TH257**

Influence of water temperature and salinity on impact of Hazardous and Noxious Substances (HNS) in the marine environment by

M. Gussow, D. Doran, C. Garcia, J. Uzyczac, M. Kirby, D. Taunton, J. Giesy, P. Thal, E. Ogburn, Environmental Standards Inc; R. Vitale, Environmental Standards, Inc.

It is recognised that Hazardous and Noxious Substances (HNS) transported at sea present a broad range of potential marine spill scenarios due to wide range of fate and effects of the many types transported in bulk through national and international waters. To improve preparedness of response and to provide better advice during marine incidents there is a need to improve our knowledge of the marine hazards/risks associated with the highest priority chemicals. Harbours and ports are typically located in transitional and coastal waters and so ship traffic is more concentrated around these areas and so potential collisions are more likely. These areas are characterised by a range of salinity from 10 - 20 ppt within estuaries up to 34 ppt in coastal and offshore waters. Similarly, seawater temperatures vary significantly with seasonality (from 6°C to 20°C in the UK). Based on these levels of salinity and temperature, preliminary studies were performed to investigate the toxicity of HNS associated with different salinities (from 20 to 30 ppt) and temperatures (10 - 25°C). Different chemicals were chosen for the tests. Aniline and zinc sulphate were chosen as they are high priority HNS Chemicals due to their relative frequent transport in bulk quantities, and zinc sulphate were also investigated as they are transported in moderate quantities, relatively frequent transport in bulk quantities, and benzalkonium chloride as a surfactant and dichlorodiphenyltrichloroethane as a disinfectant sodium hypochlorite were also investigated as they are transported in moderate quantities, have different chemical properties and modes of toxicity. Toxicity tests were performed with a micro crustacean, *Tisbe battagliai*, and two seaweeds, *Ceramium tenuicorne* and *Fucus vesiculosus*. Our results show that in most cases, chemical toxicity is positively correlated with temperature (higher toxicity with increasing temperatures) and negatively correlated with salinity (higher toxicity with increasing salinity). This means that chemical spills are likely to have more impact in the summer in temperate regions and in lower salinity coastal or estuarine areas. These are also the areas that due to the presence of cities and port and harbour facilities have higher
TH258 Using Correlations of Biological Toxicity Equivalent Quotients and Toxicity Equivalent Quotients to Derive Threshold Values for Dioxin-Like Compounds in Sediment

J.J. Díez-Juarros, RWTH Aachen University / Department of Ecosystem Analysis; M. Brinkkemper, University of Saskatchewan / School of Environment and Sustainability and Toxicology Centre; C. Gembé, Institute for Environmental Research RWTH Aachen University / Department of Ecosystem Analysis; S. Buchinger, Federal Institute of Hydrology / Department G Biochemistry Ecotoxicology; G. Reiffscheid, German Federal Institute of Hydrology; H. Hollert, RWTH Aachen University / Institute for Environmental Research Sediments can act as a sink and source of pollution in aquatic environments, particularly with respect to persistent organic pollutants (POPs) which bind to sediments and then can be released into the aquatic environment if, and when, the sediments are disturbed (e.g., dredging, floods, storm events). Among POPs, dioxin-like compounds (DLCs), which consist of a variety of contaminants that share similar structures and can bind to the arylhydrocarbon receptor (AhR) in cells, are of particular concern. In addition to chemical analyses, which are often expensive and unnecessary if the contamination is low or below threshold concentrations, measurement of the induction of ethoxyresorufin-O-deethylase (EROD) activity using the rat hepatoma cell line (H4IIE) has been identified as a potential bioanalytical screening tool for the presence of DLCs in the environment. In the presented project, the biochemical component involved the use of a 96-well plate-reader–based assay to measure EROD induction with the rat hepatoma cell line H4IIE. The micro-EROD assay can be used to determine the cytochrome p450 subfamily 1a (CYP1A)-inducing potential of a variety of substances, including extracts of sediment samples. For this project, micro-EROD assays and chemical analyses were performed on extracts of 22 sediment samples collected from wadeable bodies in Germany. We investigated the correlation of biological toxicity equivalent quotients (BEQs) determined from H4IIE micro-EROD to toxicity equivalent quotients (TEQ) determined from chemical analysis of the sediment extracts for PCDD/Fs and DL-PCBs. Correlation analysis indicated strong significant relationships between BEQs and TEQs for PCDD/Fs (r²=0.94, p<0.001) and DL-PCBs (r²=0.924, p<0.001). From these correlations, threshold values can be established and the assay used as a pre-screening tool to identify samples that would require additional chemical analyses.

TH259 Measuring bioconcentration of cationic surfactants in fish

A. Pugazhendi, C.L. Chen, Stockholm University / Department of Environmental Science and Analytical Chemistry ACES; M. McLeachlan, Stockholm University / Environmental Science and Analytical Chemistry (ACES); J.M. Armitage, University of Toronto - Scarborough / Physical and Environmental Sciences; J.A. Arnot, ARC Arnot Research & Consulting / Adjunct Professor, Department of Physical & Environmental Science/Department of Pharmacology and Toxicology; S. Kierkegaard, C.L. Chen, Stockholm University / Department of Environmental Science and Analytical Chemistry (ACES). Measuring the bioaccumulation of cationic surfactants in fish is challenging. Their disposition in water depends on both pH and alkalinity. Many cationic surfactants have a tendency to sorb extensively to surfaces, making it difficult to generate and maintain constant concentrations in aquatic water. They can also sorb extensively to the surface of fish, making it difficult to separate internal exposure from external exposure. They display a partitioning behaviour that is similar to biomolecules, making it challenging to separate them from major matrix components in fish tissue samples. They can also be toxic to fish, which constrains the concentrations that fish can be exposed to. We are currently working to define the cationic surfactant property space that is amenable to measurement of bioconcentration factors in fish. We will exploit this property space to measure the bioaccumulation behaviour of a range of cationic surfactants. This data will be used to evaluate the BIONIC model, a mechanically based model employing in vitro assay derived key input parameters (membrane-water partition coefficients and intrinsic hepatic clearance). The BIONIC model can in turn be used to estimate bioaccumulation of cationic surfactants in the property space that is not amenable to measurement. Our first experiments are conducted with sodium dodecyl sulphate (SDS) in primary, secondary, tertiary and quaternary amine range in chain length from C9 to C16. The test chemical mixture is infused continuously into the water inflow of a flow-through aquarium using a syringe pump. To determine the concentrations of the test chemicals in aquarium water, 400 µL of aquatic water is transferred with an auto-pipette to a vial containing 600 µL of methanol, and this mixture is analysed with GC/MS/MS. This method allows measurement of the high ng/L concentration range with a precision of 2-8%. Concentrations in the aquarium were maintained at a constant level for more than a week, whereby the ratio measured:nominal decreases with chain length. To determine the concentrations in fish tissue, methanol extracts are cleaned up on a weak cationic exchange SPE column followed by large-volume injection. This method allows quantification in the low ng/g range. The results of the first bioconcentration experiments will be presented.

TH260 Acetylcholinesterase inhibition: a comparison of available methods for determination of acetylcholinesterase in muscle tissue of Limanda limanda.

J. Użyczek, Centre for Environment, Fisheries and Aquaculture Science (Cefas) / Environmental and Animal Health

Acetylcholinesterase inhibition (AChE) has been used as a biomarker of the effects of organophosphate and carbamate compounds. AChE is present in most animals and is responsible for the rapid hydrolytic degradation of the neurotransmitter acetylcholine (ACh) into acetate and choline. The inhibition of AChE activity can be used to regulate the nervous transmission by reducing the concentration of ACh. When AChE is inactivated by an organophosphor or carbamate ester, the enzyme is no longer able to hydrolyse ACh and the concentration of ACh remains high. Continuous stimulation of the muscle or nerve then occurs, resulting in tetany and eventually paralysis and death. The ICES/IOC International workshop on Biological Effects of Contaminants, that took place in Bremerhaven, Germany during March 1990, provided on opportunity to test AChE inhibition as an index of marine contamination. An official ICES Technique in Marine Environmental Sciences (TIMES No.22 Biological effects of contaminants: Cholinesterase inhibition by organophosphor and carbamate compounds) is available and recommended for contaminant monitoring programmes in the marine environment. This method was published to improve and standardise the comparability between results from different laboratories and/or countries. However, the method has not been updated since 1998 and does not provide enough details on different marine species, preparation and handling of samples or storage conditions. The search continues for new monitoring tools, improvement and harmonisation of existing methods, which may be used as specific markers for contaminant effects on the safety of marine waters. For this purpose, many laboratory studies have been performed. The unique issue of Limanda limanda collected from sampling areas in the North Sea as a part of the Clean Seas Environmental Monitoring Programme (CSEMP)in the UK. The methods showed different results but verified fundamental requirements in all the procedures such as storage conditions, age of the samples or temperature dependence. More work needs to be done to standardise different approaches and combine them in a more detailed and accurate guideline. This should help to obtain precise, consistent and comparable results across the national and international laboratories and therefore provide a real evaluation of the status of the marine environment.

TH261 Environmental emission to surface water for analogous exposure path. A reflection on the matter for biocides, human and veterinary medicines.

A. Haro-Castuera, R. Carapeto García, Spanish Medicines Agency / Veterinary medicines; G. Cortés Ruíz, C. Rubio Montejano, Spanish Medicines Agency / Department of Veterinary Medicines

One emission has happened for one active substance the chain of events affecting the Environment follows its path. But how we study them depends upon the approach, dictated by legislative frames, subsequent guidance and, eventually, inertia and tradition. One remarkable example is the case of insecticides. While sharing the same active substance, different products authorized under different regulation can be applied differently. Then, to be marketed, scientific evidence of safety is mandatory. For this purpose, legislative acts are needed. The unique issue for the Environment share some principles making emphasis in different areas. Here, we will review emission paths and key risk elements as a thought starter pursuing harmonizing approaches and resource sharing between assessment schemes.

Hazard and exposure assessment of chemical mixtures: steps towards increasing the realism of chemical risk assessment (P)

TH263 Using microarthropod community assays in metal mixture testing

J. Renaud, CPE - Centre for Functional Ecology; T. Natal da Luz, University of Coimbra / Department of Life Sciences; University of Coimbra; S. Siciliano, University of Saskatchewan / Department of Soil Science; J. Sousa, University of Coimbra / Department of Life Sciences

Due of anthropogenic activities metal contaminated sites are a constant environmental concern and because of the persistence of these metal elements, will continue to be a pressing issue for many years to come. In terms of legislation and environmental risk assessment, much effort has been undertaken to understand the effects of metals and, more recently, the effects of metal mixtures. However, most legislation still focuses on single metal elements, disregarding mixtures. Similarly, research has been predominantly focused on single metals while mixture studies have mostly focused on binary and tertiary mixtures to determine metal interactions and potential synergies. However, the addition of metals is the most commonly accepted model and that considered in legislation. These studies provide valuable information on the metal mixtures but are performed with few standard test species and use mixture ratios optimized for the goal of modelling mixture interactions, which many times lack environmental relevance. In this presentation we take a different approach and test three complex five element metal mixture ratios (Pb, Cu, Ni, Zn, Co) using a natural soil microarthropod community. These metal mixture ratios were selected base on environmental and legislative relevancy, two ratios...
produced from guideline values (Canadian soil quality guideline and EU REACH PNEC values) and a ratio based on the average concentrations in a contaminated site (Sudbury) for each metal. Each mixture was tested with 11 doses in toxic units estimated from Folsomia candida reproduction EC50 for each metal in the mixture. The community results from this experiment were transformed to similarity matrices using the Bray-Curtis coefficient and used to calculate dose response curves. This approach assumes that community changes are promoted by increasing metal contamination. These constraints do not allow the estimation of microarthropod community EC values, which provide valuable insight on the adequacy of current guideline values and in developing site-specific risk assessments and remedial goals with community endpoints. Produced EC values from this simplified community experiment are currently under validation in a terrestrial model ecosystem experiment, for these same mixtures.

TH264 Alteration of stress-related and thyroid hormone related genes in zebrafish larvae after the administrations of lead acetate, and mixtures of lead acetate and BDE-209
K. Chan, The Chinese University of Hong Kong / Life Sciences; C. Leung, The Chinese University of Hong Kong / School of Life Sciences; Z. Zhou, J. Yang, The Chinese University of Hong Kong / School of Life Science
The expression profile of oxidative stress-related genes (sod1, sod2, sod3a, cc, cat, gr, gst) and thyroid-related genes (tr, tff1, dio1, dio2, nis) in zebrafish larvae was examined upon lead acetate treatments to have a complete analysis of lead toxicity on larvae development. The expression of tr, tff1, dio1, dio2, nis, sod1-1, sod1-2, sod2-1, sod2-3, ugt1a, ugt2a1, and the above oxidative stress-related genes, was also studied to unveil the impacts of co-exposure of lead acetate and BDE-209 on thyroid hormone metabolism and oxidative stress balance. Transcripts of gr and gst were induced upon both individual exposures and co-exposure, suggesting that both metals induce oxidative stress. No synergistic effects of the two chemicals at short time (48 hr) exposure to induce oxidative stress, while the possibility of these two chemicals acting synergistically to alter the gene transcription at later time points should not be ruled out.

TH265 Assessment of the toxic interaction of lanthanides on aquatic organisms
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The relevance of lanthanides (LNs), in agricultural, industrial and, especially in high-tech applications has increased in the last decades. As consequence, more LNs are expected to enter into the environment and accumulate in the ecosystem. Although no great accumulations have so far been recorded, alterations in the LNs natural-ecology of both aerobic and anaerobic soils are expected. The toxicity focus of the single elements, however, they are commonly found as a group in nature. LNs are expected to have cumulative toxic effects on organisms, owing to their similar chemical properties, but studies as mixtures, more representative of real scenarios, are required to support this hypothesis. In this research, we evaluated the toxic interactions of binary and ternary mixtures of common LNs (La, Ce, Nd) and both individual and mixture toxicities of oxidative stress and light LNs, respectively, on seven aquatic species belonging to different trophic levels. From the seven organ studied (A. fischeri, R. subcapitata, C. vulgaris, B. calyciflorus, H. incongruens, D. magna and D. rerio) potential toxic effects were observed only in five; and the inhibitory LN effects were consistently concentration-dependent only for A. fischeri, R. subcapitata and B. calyciflorus. Bioavailable LN concentrations significantly decreased during all tests and the main decline took place at the beginning of the tests, but the extent of the decrease varied across test media. Thermodynamic speciation calculation highlighted important differences: in distilled water and 1 % NaCl, LNs were predicted to occur mainly as free ions; in more complex media, LNs appeared as free forms and with complexes with both heavy metals and light metals but these species lowered with the decrease in the ionic radius, and, in detriment of these complexes, species with LCNO- and LNI(OH)+ groups increased. The two multi-toxicity approaches used in this study (concentration addition and toxic unit calculation) showed more than additive effect for the mixtures to the bacterium A. fischeri and the alga R. subcapitata; whereas less than additive toxicity was instead observed for the rotifer B. calyciflorus. Overall, the results highlight that it should not be assumed LN toxicity is additive as so far, and predicting the response of aquatic organisms exposed to mixtures should be further research to better understand their toxic interactions in real scenarios.

TH266 Predicting the chemical and biological effects of tertiary metal mixture (Ni, Cu, Cd) to aquatic plant, Lemma gibba under different dissolved organic carbon concentrations
S. Martinez, CONICET PRIET UNLU; Y. Gopalapillai, Environment and Climate Change Canada; M. Saen, PRIET CONICET, National University of Lujiang; B. Hale, School of Environmental Sciences, University of Guelph; W.D. De Mattia, CONICET-PRJET / PRIET
Toxic effects of single metals on aquatic environments are well established. In nature, organisms are exposed to a mixture of them at different bioavailability conditions. However, this situation is not always well studied. Here Lemma gibba was exposed to Ni, Cd and Zn individually and as ternary mixtures. The influence in the uptake and toxicity of dissolved organic carbon (DOC) as an environmental ligand was studied. Two sets of tests were performed.0.5 mg/L and 10 mg/L of DOC. The metal concentrations at the mixture tests were chosen by an incomplete factorial design, resulting in controls plus 20 test cases. Froud number inhibition (%FNI) and root growth inhibition (%RGI) were calculated at the end of 7-days tests. Determinations of internal dose [Mn]tiss and external dose [Mext] were also conducted for all chronic tests. Simple metal toxicity thresholds were obtained for the three metals, resulting %RGI a more sensitive endpoint than %FNI in all cases. For the test with 0.5 mg DOC/L, Cd presented the higher toxicity, based on %RGI when concentration expressed as Mext (IC25hmRNiCd = 20.8 µg/L) being 6 times more toxic than Ni and 30 times than Zn. When concentration expressed as Mext/Cd was also the more toxic metal (IC25hmRNiCd = 76.67 µg/dry weight) being 10 times more toxic than Ni and 26 times than Zn. For the test with 10 mg DOC/L, Ni was the most toxic when dose expressed as Mext, but Cd when expressed as Mext. At the end of assays, for both DOC concentrations, [Cd]tiss, [Ni]tiss and [Zn]tiss were higher in the single metal exposure compared to the mixtures. For the mixtures exposures, the %RGI responses ranged from 17 to 94 % in the lower DOC concentration test and from 15 to 97 % in the higher. Concentration addition (CA) was the appropriate model for the mixtures’ toxicity, the combinations of concentrations were used to calculate the ‘sum of toxic units’ (STU). Deviations from CA approach were observed as relative results according with the metal under analysis.

TH267 ISOLATION AND CHARACTERIZATION OF HEAVY METAL RESISTANT BACTERIA IN SOIL SAMPLES FROM MAMBILLA ARTISANAL MINING SITE, NIGERIA
O. Otuoji, federal University Wukari / Department of Biochemistry; T. Silas, federal University Wukari; A. Martins, S. Asemave, federal University Wukari / Biochemistry
Incidence of soil contamination by heavy metals is widely increasing with the spread of industries. Antimicrobial resistance and heavy metal resistance have been a major problem in many parts of the world. The Mambilla plateau has been on the increase in recent years. Therefore, the present study was aimed at characterizing and determining resistance to lead, mercury and copper by selected bacteria strains isolated in soil from Mambilla Plateau artisanal mine and to explore their bioremediation capacity. Bacteria were isolated from soil samples obtained from different locations at the Mambilla artisanal mining site. Nigeria. Five (5) bacterial isolates and M. aurora were isolated through gram-stain analysis and was used to fit the observed metal mixture toxicity data to either Mmixt or Mmixc. The concentrations of the three metals in mixture expected to result in 50% RGI or FNI were calculated by solving the regression for Cd while holding the concentrations of Ni and Zn constant for each of the 20 cases. To determine whether CA was the appropriate model for the mixtures’ toxicity, the combinations of concentrations were used to calculate the ‘sum of toxic units’ (STU). Deviations from CA approach were observed as relative results according with the metal under analysis.

TH268 The exceptions to the rule: Metal bioaccumulation in macroinvertebrates from a contaminated soil site
S. Blomlofors, R.M. Town, Systemic Physiological and Ecotoxicological Research (SPHERE), University of Antwerp / Department of Biology; R. Blust, L. Bervoets, University of Antwerp / Department of Biology (SPHERE Research Group)
Surface waters are continuously facing a variety of anthropogenic stressors, including pollution, habitat degradation, and loss of connectivity. In such complex and dynamic environments it is challenging to unambiguously establish the effects of trace metal contamination on the resident organisms. The European Water Framework Directive (WFD) obliges member states to set specific water quality
Silver nanoparticles (SNPs) are used in industrial products worldwide. Hence, there are concerns about their potential health effects. In this study, we aimed to evaluate the cytotoxicity and oxidative stress responses in human cell lines exposed to SNPs and SNCs (silver nanoparticles coated with sulfur) with different concentrations. We also investigated whether these effects are due to direct contact or dissolution of silver ions.

**Materials and Methods**

**Cell Culture**

Human cell lines (THP-1, THP-2, and THP-3) were used for the cytotoxicity assays. The cells were cultured in a medium containing RPMI 1640, 10% fetal bovine serum, and 1% penicillin-streptomycin. The cultures were maintained at 37°C in a humidified atmosphere with 5% CO2.

**Exposure Conditions**

Cells were exposed to SNPs or SNCs at concentrations ranging from 10 μg/mL to 100 μg/mL. The exposure time was 24 hours. The control group received medium without nanoparticles.

**Cytotoxicity Assays**

Cell viability was assessed using the MTT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide) assay. The absorbance was measured at 570 nm using a microplate reader.

**Oxidative Stress Assays**

ROS (reactive oxygen species) accumulation was determined using the DCFH-DA (2',7'-dichlorodihydrofluorescein diacetate) dye, which detects intracellular ROS. The fluorescence was measured at 530 nm.

**Gene Expression Analysis**

RNA was extracted from the cells using the Trizol reagent. Gene expression levels were determined using qRT-PCR (quantitative real-time polymerase chain reaction).

**Results**

- SNPs and SNCs induced a dose-dependent decrease in cell viability.
- ROS accumulation increased with increasing nanoparticle concentrations.
- Gene expression levels were altered, with significant upregulation of pro-inflammatory cytokines and downregulation of anti-inflammatory genes.

**Discussion**

Our results indicate that SNPs and SNCs can induce cytotoxicity and oxidative stress in human cell lines. The effects are concentration-dependent and may be related to direct contact with the nanoparticles or dissolution of silver ions. Further studies are needed to understand the mechanisms behind these effects and to assess their potential impact on human health.

**Conclusion**

SNPs and SNCs pose a potential risk to human health due to their cytotoxic and oxidative stress-inducing properties. Further research is needed to fully understand the mechanisms involved and to develop strategies to mitigate these effects.
a calculation of cumulative risk is needed based on active substance endpoints, risk indicators that have already been calculated for single substance ERA such as toxicity exposure ratios or risk quotients should be used to describe the cumulative risk. In a first simple step different endpoints, species and PECs in time and space can be mixed, to show that a given scenario is of low concern. After that first step, a mixture toxicity assessment would describe the cumulative risk more precisely at a given time and place for a defined species and the same endpoint.

TH274 Sublethal toxicity of pesticide mixtures on early life stages of non-target aquatic organisms
E. Rozankova, RECETOX, Faculty of Science, Masaryk University / Research centre for toxic compounds in the environment RECETOX; B. Morin, J. Cachot, University of Bordeaux / EPOC; P. Gonzalez, University of Bordeaux / UMR EPOC CNRS 5805; L. Blaha, Masaryk University, Faculty of Science / Research centre for toxic compounds in the environment RECETOX
Pesticides are widely used throughout the world in many agricultural and domestic activities. By their presence in the environment, they can have an impact on non-target organisms. Moreover, due to the persistence of some products and the formation of active metabolites, more or less complex mixtures of pesticides can be found in the environment. Thus, the aim of this study was to evaluate the effects of one herbicide (S-metolachlore and his two metabolites) and an insecticide (imidaclopride) on the embryo-larval development of two non-target aquatic organisms. These pesticides are the most abundant representatives of their groups in the Arcachon Bay in France. We were focused on environmentally relevant concentration in this Bay (1 µg/L for herbicides and 0.2 µg/L for insecticide and 2-3 higher concentrations) and their sub-lethal effects on the oyster (C. gigas), which is widely present in the Bay due to oyster farms and the zebrafish (D. rerio), chosen as a prototypical (eco)toxicology model organism. Firstly, the embryos were exposed to the separate substances, then the mixtures for 5 days (zebrafish) or 2 days (oysters). The malformations, the locomotion activity and target gene expression levels were assessed to understand the mechanisms of possible sublethal toxicity of the selected pesticides. According to our results, no malformations and no effect on locomotion activity for the tested concentrations were observed for zebrafish. On the contrary, the effects on the malformations and the locomotion activity of the oyster larvae is already induced at low concentrations of the selected pesticides. The first results of the gene expression show an overexpression of some of the selected genes of zebrafish (12S, TR-beta – known to be related to the thyroid disruption) caused by one of the metachlor metabolites. In conclusion, an indication of a novel mode of action of the chronic toxicity pesticide has been detected in zebrafish. The embryo-larval stages of oyster seem highly sensitive even to low environmental concentrations of pesticides.

TH275 Including multistress in risk assessment of pesticides. Current state of knowledge, based on a literature review and evaluation of tank mixture applications in a spraying schedule for strawberries.
P. Van der Zande, Wageningen University and Research / Agrosystems Research; G. Arts, Wageningen Environmental Research (Alterra) / Environmental Risk Assessment; W. Beltman, Alterra Wageningen UR; H. Holtermann, Wageningen University & Research / Agrosystems Research; L. Wipfler, Alterra Wageningen UR / Environmental Risk Assessment Team; J. Van de Zande, Wageningen University and Research / Agrosystems Research
This study presents a number of recent studies towards answering the question if the current product-by-product and active substance-by-active substance evaluation provides sufficient protection in the context of the authorisation of plant protection products (PPPs) in the Netherlands. This report is based on a literature review and an evaluation of tank mixture applications in a spraying schedule for strawberries. The topic of tank mixes has been identified by Cgb (the Board for the Authorisation of Plant Protection Products and Biocides in The Netherlands) as an important knowledge gap. We have quantified the environmental risk for an intensively cultivated crop with sequential applications of products and mixtures of products based on a realistic application schedule and spray drift on surface water in a ditch, the corresponding exposure profiles and the effects based on the Regulatory Assessment (RA) of the used active substances. This study shows that the actual strawberry crop scenario is not protective for invertebrates and fish in surface water. Therefore, for the risk assessment of PPPs it needs to be considered that PPPs are part of a crop protection programme and thus should be evaluated in this context. Keywords: multistress, pesticides, environmental risk, aquatic Poster presentation

TH276 MODELLING ACUTE AND CHRONIC RISKS OF PESTICIDES RESIDUES IN SOUR CHERRIES
I. Simion, Gheorghe Asachi Technical University of Iasi Romania; R. Hilhor, Gheorghe Asachi Technical University of Iasi Romania; M. Rosca, Gheorghe Asachi Technical University of Iasi Romania; M. Rădulescu, University of Iasi Romania; EPOC CNRS 5805; L. Blaha, Masaryk University, Faculty of Science / Research centre for toxic compounds in the environment RECETOX
Acute and chronic pesticide residues in sour cherries were subject to the experiments. We followed variation of environmental conditions: temperature, humidity, rainfall patterns and pesticide dissipation in time considering each treatment. The results of pesticides concentration at harvest allowed us to model the pesticides risks to human health. Based on our assessment, it appears that acute and chronic risks of pesticides residues in sour cherries are low. Sour cherries dietary intake of pesticides residues poses an acute risk for children lower than 64.6% and lower than 22.5% for adults. The highest chronic risk level reaches 2.4% for adults and 9.5% for children. Our study suggests that the risk assessment estimates are strongly influenced by age and dietary preferences.

TH277 Environmental and Human Cumulative Risk Assessment of Pesticides Using Local Monitoring Data: A Case Study from the Pucara River Basin, Bolivia
L. Herrero Noareda, University of Copenhagen / Department of Plant and Environmental Sciences; M. Álvarez Caero, Antezana Fernández, Universidad Mayor de San Simón / Facultad de Ciencias y Tecnología; N. Cedercren, University of Copenhagen / Department of Plant and Environmental Sciences
In Bolivia, pesticides are applied to the barley crop scenario is not protective for invertebrates and fish in surface water. The acute risks of pesticide residues poses an acute risk for children lower than 64.6% and lower than 22.5% for adults. The highest chronic risk level reaches 2.4% for adults and 9.5% for children. Our study suggests that the risk assessment estimates are strongly influenced by age and dietary preferences.

TH278 Developing a strategy to improve the environmental risk assessment of multi-component substances: a new HESI Emerging Issues Committee
D.T. Salvito, Research Institute for Fragrance Materials, Inc. / Department of Environmental Science; M.R. Embry, ILSI Health and Environmental Sciences Institute (HESI)
An international workshop was held in 2016 to address challenges in assessing ecological risk of complex chemical mixtures, especially for children. The detection of pesticides that caused malformations were heptachlor (banned) and dimethoate (approved). Based on these results, risk management strategies should be highly prioritized in order to reduce the risk posed to the ecosystem and the human health in the studied area. However, further studies should analyse the pesticide levels in other times of the year to see if the present results are representative. It is also necessary to analyse a higher number of pesticides which include an even representation of herbicides, fungicides and insecticides, to be sure that risk is not underestimated.
environmental risk assessment. Bridging from the workshop discussions and conclusions, a new HESI Emerging Issues Committee was formed in late 2017, with the overall mission to develop a tiered approach to UVCB and MCS ecological risk assessment. Initial objectives of this committee are to identify and develop models and methods, develop best practices and guidance, and engage with multi-stakeholder collaborative research projects. This presentation will highlight the initial goals and strategy of this multi-stakeholder, collaborative group.

**TH279 Environmental Risk Assessment of Technical Mixtures under REACH**

E. Hassold, W. Galert, German Environment Agency - UBA / IV 2.3 Chemicals; W. Dres, Federal Environment Agency (UBA) / Chemicals Analysis. The appropriate substances regulated under REACH end up in mixtures. During their processing and downstream-use substances are blended together in formulations, are reformulated for different uses and end up in a variety of products. REACH addresses the safe use of single substances in technical mixtures, but not explicitly the risks arising from joint effects and exposures of the components. In contrast to other substance-oriented regulations, not the authorities, but the registrants and down-stream users have to assess and guarantee the safe use of the registered substances, formulations and products. Recently, some attempts have been made by industry organizations with the concept of LCID/SUMI to improve the assessment and communication of safe use conditions for technical mixtures. However, essential improvements are needed. The development of sound prioritization criteria is essential for a mixture assessment. But a sole consideration of the individual constituents, 

**TH280 Natural complex mixtures: Ecotoxic behaviour, what we know and what is next?**

R. Samsera, CEHTRA SAS; N. Delpitt, Laboratoires des Pyrénées et des Landes; P. Bichere, KREATiS; J. Rivera, A. Barret, C. Durou, CEHTRA SAS; P. Chure, CEHTRA SAS / Econometrics. With June 2018 coming, the registration steps of chemicals under REACH review program come to an end but with much more to follow. But now it is time to consider the overall tasks we performed to meet that 2018 deadline. All the different classes of organic substance under REACH were considered during the Phase-in period: monoconstituents, multiconstituents, & UVCBs. Amongst these substance types, the general framework for the testing especially in ecotoxicology and environment. One of the most challenging groups was fragrances. In fragrance chemicals fall under multiple categories: natural, synthetic, mononconstituents, multiconstituent or considered as UVCBs. One group of fragrances that fell under the title of multiconstituent/UVCB were known to be particularly difficult to assess: Essential oils (EO). EO are complex mixtures, with differing but often low solubility, high volatility and are known to contain some toxic constituents such as terpenes. We met some even more complex substances in this family: gums, resinoids and concretes, sub-categories of essential oils. They were as complex as EO but their composition was mostly unknown and their physical state leads to further difficulties for ecotoxicity testing: they were (mostly) solidifying, extremely viscous resins, with a frozen honey-like texture. We tested hypotheses and performed ecotoxicology and e-fate studies on all the different classes of these substances (gums, resinoids, concretes and everything in between) to optimize our testing strategies for such compounds: we will necessitate avoiding some of these studies using alternative approaches. We will present our hypothesis and conclusions on the probable next steps for these complex substances.

**TH281 Testing chemical mixtures: how to determine the effects concentration(s)?**

G. Deviller, DERAC / TERA PRAPS HSE

When the properties of a mixture cannot be estimated from the related properties of its components then testing on the mixture is required under most chemical regulations. However, the available standard methodologies to assess the environmental fate and toxicity have been developed for single substances and are often not directly applicable to mixtures. The first issue is related to the identification of the relevant constituents to monitor during the tests (e.g. composition main constituents, bioavailable fraction...) which may differ according to the substance regulatory frame(s). Second, the development of a specific and quantitative analytical method for each relevant constituent could be technically challenging because (1) all analytical standard substances might not be available since some constituents of the mixture are produced by reaction and (2) the different chemical nature of the constituents may require different type of analytical techniques that might not be (all) available in (the same) GLP testing laboratories. Once the analytical tools have been set-up, further difficulties may arise during toxicity tests if one or more constituent's concentrations vary during the exposure time. Indeed, if the constituents are found to be all stable, then the effect concentrations (e.g. ECx or NOEC) can be based on the nominal concentration of the mixture. But, in case the constituents have different degradation patterns during the test, then the recommendation for single substances to base the effect concentrations on the measured concentration should be applied for these mixtures? This presentation is intending to discuss these issues and to bring some elements of response based on case studies.

**TH282 Deriving USEtox human non-cancer toxicity Effect factors from the REACH database for thousands of chemicals using R-Studio program**

F. Biganzoli, EU Commission Joint Research / Directorate D Sustainable Resources Bioeconomy Unit; L. Ceriani, M. Fuart-Gatnak, M. Pavan, S-IN Soluzioni Informatiche Srl; A. Gissi, European Chemicals Agency; E. Saouter, EU Commission JRC / Sustainable Assessment Unit

The Product and Organization Environmental Footprint (PEF/OEF) methods form a core part of the European Commission (EC) Recommendation “on the use of common methods to measure and communicate the life cycle environmental performance of products and organisations” (2013/179/EU). Based on these methods, the potential impact on humans and ecosystems of chemicals emitted during the life cycle of a product is assessed via the USEtox model. To this aim, various inputs are required to model its life cycle impact: (1) data input for a chemical. Focusing on human health (HH), the EC Joint Research Centre has recently obtained from the European Chemicals Agency (ECHA) genotoxicity and repeated dose toxicity (RDT) data (41’381 test results, as of March 2017) generated under the REACH Regulation. Based on these data, selection criteria were defined to automatically derive non-cancer HH toxicity effect factors (using R-studio) for thousands of chemicals in USEtox. Genotoxicity data were not retained in the assessment, being associated with qualitative outcomes, and rules for cancer HH effect factors were not derived, since the USEtox ED50, and cancer-TD50 endpoints are not commonly provided under REACH. According to the USEtox methodology, specific fields of the REACH data, included in the RDT endpoint study records (ESRs) via the oral and inhalation route, were used to define selection criteria for non-cancer HH effects, in particular: reliability, adequacy, type of information, test guideline, GLP, species, duration of exposure, route of administration, effect level qualifier, effect level, unit, effect level based on, basis for effect levels. A tiered approach for selecting good quality data was also proposed, based on four quality levels, where studies of the highest quality (key studies, Klimisch 1/2) were included in the first two levels.

**TH283 Deriving USEtox aquatic freshwater toxicity Effect factor from OpenFoodTox database (EFSAs) using R-Studio program**

E. Saouter, EU Commission JRC / Sustainable Assessment Unit; F. Biganzoli, EU Commission Joint Research / Directorate D Sustainable Resources Bioeconomy Unit; L. Ceriani, S-IN Soluzioni Informatiche Srl; J. Dorne, European Food Safety Authority EFSAs / Scientific Committee and Emerging Risks Unit Department of Rival Casamassima; J. Richardson, EFSA / AMU; D. Versteeg, EcoStewardship LLC; F. Biganzoli, EU Commission Joint Research / Directorate D Sustainable Assessment Unit; L. Ceriani, S-IN Soluzioni Informatiche Srl; J. Dorne, European Food Safety Authority EFSAs / Scientific Committee and Emerging Risks Unit Department of Rival Casamassima; J. Richardson, EFSA / AMU; D. Versteeg, EcoStewardship LLC; F. Biganzoli, EU Commission Joint Research / Directorate D Sustainable Assessment Unit; L. Ceriani, S-IN Soluzioni Informatiche Srl; J. Dorne, European Food Safety Authority EFSAs / Scientific Committee and Emerging Risks Unit Department of Rival Casamassima; J. Richardson, EFSA / AMU; D. Versteeg, EcoStewardship LLC; F. Biganzoli, EU Commission Joint Research / Directorate D Sustainable Assessment Unit; L. Ceriani, S-IN Soluzioni Informatiche Srl; J. Dorne, European Food Safety Authority EFSAs / Scientific Committee and Emerging Risks Unit Department of Rival Casamassima; J. Richardson, EFSA / AMU; D. Versteeg, EcoStewardship LLC; F. Biganzoli, EU Commission Joint Research / Directorate D Sustainable Assessment Unit; L. Ceriani, S-IN Soluzioni Informatiche Srl; J. Dorne, European Food Safety Authority EFSAs / Scientific Committee and Emerging Risks Unit Department of Rival Casamassima; J. Richardson, EFSA / AMU; D. Versteeg, EcoStewardship LLC; F. Biganzoli, EU Commission Joint Research / Directorate D Sustainable Assessment Unit; L. Ceriani, S-IN Soluzioni Informatiche Srl; J. Dorne, European Food Safety Authority EFSAs / Scientific Committee and Emerging Risks Unit Department of Rival Casamassima; J. Richardson, EFSA / AMU; D. Versteeg, EcoStewardship LLC; F. Biganzoli, EU Commission Joint Research / Directorate D Sustainable Assessment Unit; L. Ceriani, S-IN Soluzioni Informatiche Srl; J. Dorne, European Food Safety Authority EFSAs / Scientific Committee and Emerging Risks Unit Department of Rival Casamassima; J. Richardson, EFSA / AMU; D. Versteeg, EcoStewardship LLC; F. Biganzoli, EU Commission Joint Research / Directorate D Sustainable Assessment Unit; L. Ceriani, S-IN Soluzioni Informatiche Srl; J. Dorne, European Food Safety Authority EFSAs / Scientific Committee and Emerging Risks Unit Department of Rival Casamassima; J. Richardson, EFSA / AMU; D. Versteeg, EcoStewardship LLC; F. Biganzoli, EU Commission Joint Research / Directorate D Sustainable Assessment Unit; L. Ceriani, S-IN Soluzioni Informatiche Srl; J. Dorne, European Food Safety Authority EFSAs / Scientific Committee and Emerging Risks Unit Department of Rival Casamassima; J. Richardson, EFSA / AMU; D. Versteeg, EcoStewardship LLC; F. Biganzoli, EU Commission Joint Research / Directorate D Sustainable Assessment Unit; L. Ceriani, S-IN Soluzioni Informatiche Srl; J. Dorne, European Food Safety Authority EFSAs / Scientific Committee and Emerging Risks Unit Department of Rival Casamassima; J. Richardson, EFSA / AMU; D. Versteeg, EcoStewardship LLC; F. Biganzoli, EU Commission Joint Research / Directorate D Sustainable Assessment Unit; L. Ceriani, S-IN Soluzioni Informatiche Srl; J. Dorne, European Food Safety Authority EFSAs / Scientific Committee and Emerging Risks Unit Department of Rival Casamassima; J. Richardson, EFSA / AMU; D. Versteeg, EcoStewardship LLC; F. Biganzoli, EU Commission Joint Research / Directorate D Sustainable Assessment Unit; L. Ceriani, S-IN Soluzioni Informatiche Srl; J. Dorne, European Food Safety Authority EFSAs / Scientific Committee and Emerging Risks Unit Department of Rival Casamassima; J. Richardson, EFSA / AMU; D. Versteeg, EcoStewardship LLC; F. Biganzoli, EU Commission Joint Research / Directorate D Sustainable Assessment Unit; L. Ceriani, S-IN Soluzioni Informatiche Srl; J. Dorne, European Food Safety Authority EFSAs / Scientific Committee and Emerging Risks Unit Department of Rival Casamassima; J. Richardson, EFSA / AMU; D. Versteeg, EcoStewardship LLC; F. Biganzoli, EU Commission Joint Research / Directorate D Sustainable Assessment Unit; L. Ceriani, S-IN Soluzioni Informatiche Srl; J. Dorne, European Food Safety Authority EFSAs / Scientific Committee and Emerging Risk...
available for about 451 chemicals. After selecting the appropriate data, species geometric means have been calculated for each taxonomic groups. The following final calculation have been performed for thousands of chemicals: - Acute and Chronic species geometric means with standard deviation and number of individual test available per species - Arithmetic average of all the log of the species geometric mean with standard deviation and count of species as well as count of SSD group for each chemical and Chronic species geometric means with standard deviation Comparison of the chemical toxicity based on the above calculations and chemical ranking based on GHS and CLP.

TH284 Bioassays for assessing effects of overall risk from food contact materials K. Esfandiari, Fase Foundation; J. Muncke, Food Packaging Forum Foundation / General Management

Food contact articles (FCAs) are made from highly diverse materials, and they are chemically complex. FCAs can transfer their chemical constituents, the so-called food contact chemicals (FCCs), into foods. Exposure to FCCs is assumed to be highly relevant in the context of human exposure to (synthetic) chemicals. To assess the risk to human health from chronic ingestion of FCCs, basic information on migrating chemicals must be available, such as their chemical identity.

This, however, is often not the case for all migrating FCCs, especially the non-intentionally added substances (NIAS), as some or most NIAS typically remain unknown, depending on the type of FCA. Furthermore, the current approach to chemical risk assessment is focused on single substances, while it is known that many FCCs migrate simultaneously, forming the ‘overall migrant’ and resulting in typical and predictable mixture exposure scenarios. One alternative approach to estimating chemical hazards of FCAs is to assess biological effects of the overall migrate. In addition to assessing mixture toxicity this approach also includes effect- assessment for unknown NIAS which otherwise remain unassessed.

We review this approach, discuss benefits and disadvantages, and highlight future research needs.

TH285 A unique index to characterize the global noxiosiness of stable and radioactive substances for both human health and ecosystems K. van Neck, Eefra, Institut de Radioprotection et de Sûreté Nucléaire / SRTE; R. Gilbin, Institut de Radioprotection et de Sûreté Nucléaire / PAF; S. Reygrobeltel, Institut de Radioprotection et de Sûreté Nucléaire / PAF-ENV/SRT2; J. Garnier-Laplace, Institut de Radioprotection et de Sûreté Nucléaire / PAF-ENV

Inspired by methods and tools developed in the field of life cycle analysis (LCA), we are developing an index to assess the hazard to human health and environment due to stable and radioactive substances. Six impact categories were considered: human cancer and non-cancer effects on one hand, and ecotoxicity on the other hand, both considering chemotoxicity and/or radiotoxicity. For ecosystems, a comparative toxic unit has already been defined from which we derived our noxiosiness index. It is based on the concept of Potentially Affected Fraction (PAF), used here as a damage indicator at the ecosystem level. This concept expressed initially the toxic pressure on the environment due to one chemical. It has been enlarged to mixtures of substances as multi-substances PAF (ms-PAF), and yet applied to a mix of stable and radioactive substances. Combining ecotoxicity data and a simplified model of exposure of fauna and flora, we proposed a chemotoxicity index and a radiotoxicity index, which definitions ultimately allow the derivation of a single index. According to acknowledged practices in LCA and corresponding available data, we suggested declining this approach to human health, taking into account exposures resulted from both ingestion and inhalation pathways. This led to eight basic indices, which may be aggregated on substances, effects categories or exposure pathways to produce intermediate indexes. The principle of additivity that underlies the whole proposed approach may authorize their complete aggregation in order to produce a unique index also for human health. Different source terms may be then easily directly compared in terms of human and ecological noxiosiness.

TH286 Solution-focused application of mixture modelling and chemical footprints M.C. Zigg, RIVM / Centre for Sustainability Environment and Health; J. van Gils, DELTARES; A. van Wezel, KWR Watercycle Research Institute / Chemical Water Quality and Health; D. De Zwart, DeZ Ecotox / Centre for Sustainability Environment and Health; D. van de Meent, Association of Retired Environmental Scientists AES / Environmental Science; L. Posthuma, RIVM / Centre for Sustainability Environment and Health

Status reports of the Water Framework Directive suggest many cases of non-compliance according to formal criteria for Good Ecological and Good Chemical Status as well as for River-Basin Specific Pollutants. This signals problems for both the assemblages of species and the functions we try to protect. Multiple stressors, including 100k chemicals and their mixtures, are the causal agents. In the FP7 project Solutions, a modelling train is developed with the purpose to support derivation of water quality management plans that provide best value for money regarding chemicals and their mixtures, i.e., help to identify the largest potential risk reduction per euro spent. Thereby, the challenge is to focus on those chemicals that matter most, which can result in prioritization to sites (where are largest impacts to be expected), to times (when are largest impacts to be expected, e.g., crop-growing season & pesticides), to affected species groups (which species [groups] are most sensitive to the present impact) and to compound (which chemical [groups] contribute most to local impacts). Collaboration with the FP7-project MARS (on multiple-stress effects on ecological status) forms a multi-stress, multi-chemical inchunycolux. In SOLUTIONS, the modeling train will result in chemical footprints (ChF). Chfs were developed to summarize and communicate predicted mixture risks in simple units. In the current presentation, ChFs are evaluated in terms of the net contributions to predicted mixture impact of emissions in one region, e.g., a sub-catchment, on the mixture toxic pressure in the total river downstream, including the estuary. Hence, it exists potential transfer from one ecological level to another which clarifying the information that can be obtained by single-chemical risks, chemical footprints indicate which combination of chemicals and locations require priority for abatement. We present ChF-analysis results for the river Rhine. The river basin was divided in more than 800 sub-basins and chemical footprint information resulting from the emissions of a large amount of chemicals are derived from hydrology driven spatially and temporally explicit modelling for the whole catchment. Subsequently, abatement priorities are proposed, based on the ChF results, that act on the most severe combinations of chemical and region of emissions. The effects of local risk management are expressed in reduced toxic pressure in all downstream sections of the river.

TH287 One-week observation of phthalate metabolites in urine from 12 Korean adults: Exposure levels, profiles, and source identification J. Lim, Hanyang University; S. Kim, Seoul National University Graduate School of Public Health; I. Lee, Seoul National University / Department of Environmental Health Sciences; A. Lee, Soonchunhyang University; S. Kim, Y. Kho, Eulji University; Y. Park, SEOG CHUN HYANG UNIVERSITY; J. Park, Seoul National University / Center for Natural Sciences; K. Choi, Seoul University / Environmental Health Sciences; H. Moon, Hanyang University / Marine Sciences and Convergent Technology

The esters of phthalic acid (phthalates) are representative endocrine disrupting chemicals (EDCs) to cause a variety of adverse health effects to humans. Phthalates have been primarily used as additives of the plastic products, cosmetics and personal care products (e.g., DEP). Phthalates are metabolized and eliminated in urine within few hours after human exposure. Due to a short half-life of phthalates, the urinary phthalate metabolites are utilized as an indicative for integrative exposure to phthalates from multiple sources and pathways. In this study, all of the urine samples for 7 days (n=401) were collected from 12 adults to identify the identification of exposure levels, fraction profiles and exposure sources for different population groups. Eighteen phthalate metabolites include mono-ethyl phthalate(MEP), mono-ethyl phthalate (MEP), mono(2-ethyl-5-octoxyl) phthalate (MEHP), mono(2-ethyl-5-hydroxyxylo) phthalate (MEHP), mono-(2-carboxymethylxylo) phthalate (MCMPH), and mono-(2-ethyl-5-carboxypentyl) phthalate (MECPP) were analyzed. Among 18 phthalate metabolites, MEHP, MEOPH, MCMHP, MECMP, MBP, MEP, and MBzP were detected in all almost of the urine samples (detection rate >97%). However, MCHP, MiPp, MiNP, MOP, and MEP were rarely detected in all of the urine samples (<10%). Total concentrations of phthalate metabolites ranged from 3.12 to 6300 ng/mL with a median concentration of 104 ng/mL. Five DEHP metabolites were measured and expressed as the highest concentration (mean: 63 ng/mL). MiNP (median: 8.4 ng/mL), MBzP (6.8 ng/mL) and MEP (5.2 ng/mL) showed relatively higher concentrations than other phthalate metabolites. Our finding suggests the highest burden of DEHP from multiple sources. In the present study, we defined the peak showing the concentration higher than summation of average and double values of standard deviation as a specific source input associated with phthalate exposure. Tracking the exposure source of phthalates suggests that the major contribution of the phthalates exposure pathways was different depending on chemical properties (e.g., molecular weight) and usage of phthalates. The exposure of lower-molecular-weight phthalates such as DEP and DMP was associated with the consumption of cosmetics and personal care products, whereas the urinary DEHP exposure levels varied with the dietary intake. The present study provides an important information for intervention study to reduce phthalates from humans.

TH288 Integrating chemical monitoring data with high-content effects data to prioritize contaminant and hazards in chemical mixtures D. Martinez-Leo, University of St. Thomas / Biology; A.C. Mehinto, Southern California Coastal Water Research Project / Toxicology; N. Vinas, US Army Engineer Research and Development Center; A. Schroeder, University of Minnesota-Crookston / Math, Science and Technology; E.M. Curran, University of St. Thomas; C. Lai, University of St. Thomas / School of Engineering; Y. He, University of St. Thomas / School of Engineering; M.L. Ferrey, Minnesota Pollution Control Agency / Environmental Outcomes

Determining ecological risks associated with exposures to complex chemical mixtures, the so-called chemical footprints (ChFs), are developed to summarize and communicate predicted mixture risks in simple units. In the current presentation, ChFs are evaluated in terms of the net contributions to predicted mixture impact of emissions in one region, e.g., a sub-catchment, on the mixture toxic pressure in the total river downstream, including the estuary. Hence, it exists potential transfer from one ecological level to another which clarifying the information that can be obtained by single-chemical risks, chemical footprints indicate which combination of chemicals and locations require priority for abatement. We present ChF-analysis results for the river Rhine. The river basin was divided in more than 800 sub-basins and chemical footprint information resulting from the emissions of a large amount of chemicals are derived from hydrology driven spatially and temporally explicit modelling for the whole catchment. Subsequently, abatement priorities are proposed, based on the ChF results, that act on the most severe combinations of chemical and region of emissions. The effects of local risk management are expressed in reduced toxic pressure in all downstream sections of the river.
the solutions. A limitation of these is that they typically do not provide insight into which chemicals are causing the observed biological responses. Utility of methodologies that integrate chemical monitoring with bio-effects data to prioritize chemicals and hazards in complex mixtures will be discussed. More specifically, outcomes of a Minnesota streams case study are used to critically evaluate approaches where: 1) prior knowledge regarding toxicity of detected chemicals is combined with empirical, in situ bio-effects assessment, and 2) where in situ chemical and occurrence data and in situ bio-effects data are integrated directly (without the prior knowledge of toxicity of individual chemicals). Samples from 50 randomly selected locations in Minnesota were analyzed for 146 chemicals of emerging concern. Concurrently, at 10 sites, exposures of fathead minnows to stream water were conducted (48h, custom 60K feature microarray platform, liver, NanoString technology; chemistry and public bio-effects data for individual chemicals were integrated to prioritize chemicals and predict biological targets of detected chemicals for each site. Partial least-squares (PLS) regression and association rule learning (AR) were used to identify associations between in situ chemistry and in situ transcriptomic effects. At most sites, both prior knowledge-based predictions and fish transcriptomics, indicated activation of estrogen receptor alpha and peroxisome proliferator-activated receptors; their predicted chemical initiators were bisphenol A, caffeine, carbamazepine, and triclosan. Some chemicals (triclosan) were indicated by both knowledge-supervised and direct data integration approaches, but iodamold (detected at 78% of MN sites) and metformin were only indicated by PLS and AR. Estrogenic effects remain of special concern as all methodologies indicated disruption or estrogen receptor signalling. Collectively, additional toxicants is gasoline from noncurrent cars. These three health danger pollutants areas were found: abnormal high levels; lower clearance via renal; higher excretion. These health effects data are integrated directly into chemical data, wide exposure, as monitoring data can be used to evaluate model predictions and refine parameterization while modelling provides feedback to improve the representativeness of sampling. This method enables a more detailed analysis of the key sources of uncertainty and variability at each step of the modelling framework (i.e., exposure, effluent and river concentrations). Further work to understand the uncertainties in both monitoring and modelling will be carried out in a monitoring campaign in November 2017. TH291

Assessment; etc). Numerous health disorders (in children and pregnant women) of immunity, genetic, endocrine system. The old environment pollution by obsolete gasoline. TH292

Mycosporin experiment evidences complex responses of biofilm communities along a gradient of chemical pollution 1. sabater, CSIC-IDAEA / Department of Environmental Chemistry; A. Ginebreda, Institute of Environmental Assessment and Water Research IDAEA CSIC / Department of Environmental Chemistry; O. Pereda, University of the Basque Country; F. Romero, ICRA Catalain Institute for Water Research; D. Barcelo, IQAB-CSIC / Department of Environmental Chemistry. Freshwater ecosystems are receivers of chemical pollution, which has been recognized as one of the major causes of river impairment. Wastewater treatment plants (WWTP) are point sources of contaminants in natural receiving waters but information about their effects in the ecosystems is still scarce. Ecosystems are known to react to any environmental change by initiating a series of ecological chain of events to recover its altered state. In the current research leading to this study, we seek to determine how robust and resilient is an ecosystem to WWTP effluents using a mycosporace experiment which have been revealed as particularly convenient tool to study biological communities’ responses. We exposed twenty-four artificial streams to a range of WWTP effluent concentrations, from no effluent to pure effluent and under controlled conditions of light and water flow during 34 days, followed for 22 days of recovery. Further no effluent conditions. We analysed river biofilm inhabiting in sediments and cobbles surface because of its major role in ecosystem functioning. To assess impact and recovery we measured Chlorophyll-a content, Chlorophyll-a fluorescence measurements, extracellular enzyme activities (APA and LAP), algal community and metabolism at weekly scale. Pollution load associated to the WWTP effluent was characterized analyzing physical-chemical parameters (pH, dissolved oxygen, conductivity and temperature), nutrients, organic matter, heavy metals and microcontaminants in the water phase. Our preliminary results indicate a complex response of stream ecosystem functioning in front of the WWTP effluent. We notice changes on the system balance and the final return to equilibrium. Acknowledgements - The research leading to these results has received funding from the European Communities 7th Framework Programme under Grant Agreement No. 603629-ENV-2013-6.2.1-Globaqua

TH292

Risk assessment of chemical mixtures in the Erft river basin

Mixture toxicity was assessed using extensive chemical monitoring data from the river Erft, its tributaries, and ten municipal waste water treatment plants (WWTPs) draining effluents into the Erft river. Toxic Unit (TU) was calculated assuming concentration addition and using acute toxicity endpoints for algae, macrophytes, daphnia and fish. Substances with high TU as well as the taxonomic groups displaying highest added up TU response (SUM TU) were determined. The chemical inventories of WWTP effluents were analysed to gather information on non-decals i.e. potentially ecotoxicologically relevant substances which are present in surface waters in concentrations below the analytical limit of detection (LOD). Additionally, single substance risk assessment was performed by determining risk quotients (environmental concentration/PNEC). Risk quotients larger than one indicate a possible risk for aquatic organisms. SUM TU were mainly

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explained by few compounds which varied between sampling sites and dates (e.g. seasonal use of pesticides). Overall, WWTPs increased mixture toxicity in the receiving surface waters. For most samples highest SUM TU could be calculated for macrophytes and algae. As a substance highly toxic for algae Triclosan generated high TU. It was detected in nearly all WWTP effluents but in surface waters it was only rarely present in concentrations above LOD. Triclosan can be considered to be a risk factor for the aquatic organisms. In waterbodies strongly influenced by WWTP discharges Diclofenac and Busprofen were nearly ubiquitous and caused high chronic toxic stress to fish. It was concluded that a combination of single substance risk assessment and mixture toxicity assessment is a suitable tool to evaluate complex monitoring data. Monitoring of substances with high TU (contributing mainly to mixture toxicity) could help to identify surface water for a more extensive monitoring and support specific management planning.

TH293 Assessing groundwater toxicity of emerging contaminant mixtures M.D. Pavlički, University of Aveiro / Department of Biology; J.F. Mousinho, University of Aveiro / Department of Biology and CESAM; A.R. Silva, University of Aveiro / Depart. of Compost & CESAM; S. Goulart, Dettaglia and Cremona & CESAM - University of Aveiro / Department of Biology and CESAM; R. Morgado, University of Aveiro / Department of Biology and CESAM; S. Loureiro, Universidade de Aveiro / Biology

Groundwater is one of the most important natural resources, as globally it comprises the primary available source of freshwater. Groundwater aquifers consist in an important drinking source in many parts of the world and a point of supply for irrigation in agriculture, among others. Additionally, groundwater aquifers are considered valuable in sustaining ecosystems’ health and functioning. The Groundwater Directive (2006/118/EC) was created to protect groundwater bodies from contamination but to date it does not consider a diverse array of emerging contaminants in great quantities by society. These emerging contaminants can often occur in mixtures rather than alone, therefore understanding and predicting the toxicity of such complex mixtures, will eventually lead the way to developing new strategies for setting adaptations in regulations. Additionally, adapting surface water protocols to groundwater contamination scenarios might lead to erroneous results due to water different composition. The present work was performed in the context of the European Research Project WEWORKS2014 ERA-NET focusing on developing new management strategies to sustainably exploit groundwater resources. A thorough identification of emerging contaminants took place in two well-characterized case-studies, the Bologna and Cremona aquifers. For that, four priority contaminants identified in the two aquifers were chosen as model chemicals and synthetic water was built to mimic their patterns from the two aquifers. The University of Aveiro / Department of Biology and CESAM investigated the acute toxicity of complex mixtures in these synthetic groundwaters was tested in Daphnia magna and deviations from the Concentration Addition reference model were assessed. Based on this step, the toxicity of three emerging compounds and their mixtures (binary and ternary) were assessed in the two synthetic groundwaters using adapted standardized protocols for Daphnia magna (OECD 202) and Danio rerio (OECD 203). A use model was employed to simulate the mixing of binary mixtures used to predict the effects of the ternary mixtures. Deviations from the Concentration Addition reference model indicate interaction between the contaminants in D. magna and D. rerio.

TH294 Mixture effects of Dibutyl phthalate and Sodium dodecyl sulphate on a mesozooplankton community from the Swedish west coast C. Jönander, University of Gothenburg; I. Dahlöf, University of Gothenburg / Biological and Environmental Sciences; T. Backhaus, University of Gothenburg / Department of Biological and Environmental Sciences

In coastal ecosystems emerging contaminants are not present on their own but in mixtures. A chemical monitoring survey detected more than 60 organic compounds along the west coast of Sweden in 2012, many of which exceeded their environmental thresholds. A total of 33 chemicals was found to co-occur in the water column of the coast of Stenungsund, an area with multiple harbours and home to Sweden’s biggest chemical industry cluster. Dibutyl phthalate (DBP) and Sodium dodecyl sulphate (SDS) were among the highest risk substance (concentration / ecotoxicity). We therefore investigated their single substance and mixture toxicity to natural mesozooplankton communities, which constitute an important link between primary producers and higher trophic levels like fish. Structurally diverse communities generally possess a large resilience capacity, and it is thus essential to identify sensitive species and structural changes caused by chemical exposure. Potentially, structural changes on this level could indirectly affect even higher levels of biological complexity. We used copepod egg production, hatching success and feeding rate as our primary ecotoxicity parameters, complemented with dead/alive staining of zooplankton with neutral red after the exposure. Additionally, we analysed the community structure before and after chemical exposure by image analysis, comparing images of the exposed samples and untreated controls to a manually classified reference library of mesozooplankton taxa. Single substance experiments show toxic effects on the zooplankton communities by decreasing copepod egg production and hatching success in a concentration-dependent manner, with first effects becoming visible at concentrations of 0.20 μmol/L (DBP) and 0.32 μmol/L (DBP), respectively. The quantification of structural endpoints as well as toxic stress experiments are currently (Nov. 2017) ongoing and will be presented on the poster.

TH295 Analysis of the mixture toxicity burden in 17 Rivers in North Eastern Australia - Implications for the Great Barrier Reef. E. Spilsbury, University of Gothenburg / Dept of Biological and Environmental Sciences; M.S. Warne, Coventry University / Centre for Agroecology, Water and Resilience; T. Backhaus, University of Gothenburg / Department of Biological and Environmental Sciences

The Great Barrier Reef (GBR) is a protected ecosystem, listed as a UNESCO World Heritage site since 1981. It runs for approximately 2300 km along the coastline in north-eastern Australia. A total of thirty-five major river basins discharge to the GBR and many transport large loads of pesticides, suspended sediments, nutrients from agricultural land. Over the past 6 years an extensive program has been conducted by the Queensland Government to monitor concentrations of 51 pesticides and their breakdown products in 17 rivers that discharge to the GBR. To explore the potential impact of these pollutants on the GBR we analysed the toxicity risk posed by individual pesticides and their mixtures. Australia currently does not have water quality guidelines for 17 of the 38 pesticides detected. For those, we calculated ecotoxicity thresholds using a simplified version of the Australian methodology for determining water quality guideline values, based on species-sensitivity distributions. In all rivers, multiple pesticides were routinely detected at concentrations higher than their level of reporting. All rivers had at least one sample where the combined toxicity was greater than 1 toxic unit (TU), i.e. exposure situations where the total pesticide concentration exceeded acceptable levels. In a number of rivers more than 50% of samples had a combined toxicity greater than 1 TU. Average TUs per river ranged from 13.47 to 0.10, with substantial fluctuations over the seasons but without clear trends between years. The patterns indicate that specific events such as severity of wet/dry seasons and cyclone events impact the combined toxicity found. We also found land use patterns affected the combined toxicity in the river ecosystems. In each of the rivers, 90% of the expected mixture toxicity was caused by only between 2 and 6 pesticides, although the individual pesticides that dominated the combined toxicity differed between rivers.

TH296 Physiological and transcriptomic responses in the tropical coral Stylophora pistillata to inorganic sunscreen exposure. A. Tagliati, Heriot Watt University / Institute of Life and Earth Sciences; S.J. Bellwood, University of Queensland; T.F. Fernandes, Heriot-Watt University / Institute of Life and Earth Sciences; M.S. Warne, Coventry University / Centre for Agroecology, Water and Resilience

Australia currently does not have water quality guidelines for 17 of the 38 sunscreen components in it. A series of short-term (5 days) experiments were carried out to test effects of these sunscreens on corals, by studying coral polyp morphology, photosynthesis rate, bleaching and global gene expression. The expression of genes involved in thermal stress (HSP70), carbon acquisition (intra and extracellular carbonic anhydrase) and calcium and ATP exchange (CA-ATPase) were also analysed to characterize Stylophora pistillata transcriptomic response to sunscreen exposure. Results from this work will be presented at SETAC Europe 2018, 28th Annual Meeting.
The ecological risks of these mixtures are however poorly understood. In this study, the toxicity of single and mixture of six major human antibiotics from different classes; amoxicillin (AMO), oxytetracycline (OXY), clarithromycin (CLA), meropenem (MER), ciprofloxacin (CIP), cephalaxin (CEP) to the blue green algae, Anabaena flos-aquae was assessed. All antibiotics showed high toxicity to the cyanobacteria with EC50 concentration ranging from 0.001 to 0.08 mgL\(^{-1}\) (CLA, 0.001 mgL\(^{-1}\), CEP, 0.003 mgL\(^{-1}\), CIP, 0.008 mgL\(^{-1}\), OXY, 0.006 mgL\(^{-1}\), MER, 0.02 mgL\(^{-1}\) and AMO, 0.03 mgL\(^{-1}\)). Use of these toxicity data alongside predictions of surface water concentrations, using simple models, resulted in risk characterisation ratio values of 30.2, 2.5, 2.4, 1.9, 1.4 and 0.2 for AMO, OXY, CLA, MER, CIP and CEP respectively, suggesting that five of the six compounds may be adversely affecting the aquatic environment. The mixtures toxicity studies are ongoing but for the data available; these will be used to evaluate the concentration addition (CA) and independent action (IA) for estimating the mixture toxicity. The best performing model will then be used alongside exposure modelling approaches to explore the risks of mixture for different scenarios.

**TH298**

Exposure to mixtures of Persistent Organic Pollutants (POPs) can inhibit the transactivation activities of Aryl hydrocarbon Receptor (AhR) in vitro Q.T. Doan, Université de Liège (ULiege) / Département des Sciences des Denrées alimentaires; M. Muller, University of Liege / GIGA-R. Laboratory for Organogenesis and Regeneration; H. Berntsen, NMBU-School of Veterinary Science / Department of Production Animal Clinical Sciences; K. Zimmer, NMBU-School of Veterinary Science / Department of Basic Sciences and Aquatic Medicine; S. Verhaegen, E. Ropstad, NMBU-School of Veterinary Science / Department of Production Animal Clinical Sciences; L. Connolly, Queens University of Belfast / Institute for Global Food Security, School of Biological Sciences; M. Scippo, University of Liege / Department of Food Science, FARAH. AhR antagonism is an important factor for the persistence of POPs in different species. Here we aimed to investigate the possibility of AhR antagonism involved in an organism’s response to POPs as xenobiotics. In this study, different luciferase reporter cell lines (rat hepatoma H4IIE, human mammary gland carcinoma T47-D and human hepatoma Hep G2) were used to screen AhR transactivation activities (i.e. agonistic and antagonistic) of 29 compounds listed as POPs under the 2001 Stockholm Convention. Their mixture, prepared according to the concentration of each of the compounds found in human blood, was also tested for the same reasons. We show that these compounds have species- and tissue-specific effects and that the rat cells DR (Dioxin responsive)-H4IIE are more sensitive than the two human cell lines (DR-T4-D and DR-Hep G2). Only 6 out of them showed AhR agonistic activities. PBDE-153, PBDE-154, PCB-138, and PCB-118 were able to activate the AhR in H4IIE cells only, γ-HCH was active in DR-T4-D only, while PBDE-99 was found to be an AhR agonist in both cell lines. No agonistic effect was seen for DR-Hep G2. In contrast, 19 out of the 29 compounds showed AhR antagonistic activities in DR-H4IIE, while 10 and 6 compounds displayed AhR antagonistic activities in DR-T4-D and DR-Hep G2 cells, respectively. Not surprisingly, the mixture of the 29 compounds also showed an AhR antagonistic action in all cell lines. In this study, AhR inhibition was observed with concentrations of the POP mixture corresponding to 75 times the blood level and above, which could be practically reached in humans after a food contamination incident. The IC50 for the POP mixture was 262.6 ± 104.6 times the background blood level, which corresponds to an interpolated antagonistic equivalent of 0.165 mg L\(^{-1}\). By using the isobole coefficient of the mixture is 0.3 (< 1) according to additive mixture effect model. This indicates that AhR antagonistic actions are significantly enhanced in real mixtures.

**TH299**

Ecotoxicity of biofuel-mixture DbNE and 1-Octanol on aquatic organisms Danilo rorio and Daphnia magna M.D. Esser, Institute for Environmental Research WRTH Aachen; S. Heger, Institute for Environmental Research, WRTH / Institute for Environmental Research; M. Du, Institute for Environmental Research, WRTH Aachen / Department of Ecosystem Analysis; H. Hollert, RWTH Aachen University / Institute for Environmental Research; A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics. The worldwide demand for fuels is increasing, but currently used fuels are based on fossil resources. A possible alternative for diesel is the biofuel mixture of 1-Octanol (1-Oct) (80%) and Di-n-Butyl Ether (DnBE) (20%). These fuels are based on the raw material lignocellulose and Di-n-Butyl-Neutral. However, there is no literature on this mixture impact on the environment contamination, especially for aquatic ecosystems. This study focuses on the ecotoxicological evaluation of this mixture and the investigation on a possible interaction of the two substances. Acute embryotoxic and developmental effects were investigated in the fish embryo toxicity test (FET) with Danio rerio (OECD 216). The acute immobilization assay (OECD 202) was performed to determine the effect of single exposure. To interpret the results for possible interactions between the two substances, the investigation of DnBE and 1-Octanol as single substances was necessary. In the acute immobilization test, the LC50 values were 14.7 mg/L for 1-octanol and 17.3 mg/L for DnBE. Both biofuels led to teratogenic and lethal effects in the FET (LC50, DnBE: 24.7 mg/L; LC50, 1-Octanol: 11.3 mg/L). Especially in the study of DnBE was a low hatching rate, while embryos were often observed at the pericardium of the developing larva. Testing the mixture in the FET revealed a LC50 of 14.7 mg/L. The acute immobilization test resulted in a EC50 of 25.6 mg/L. The determined EC50/LC50 values in both bioassays suggest an additive mode of action of the compounds. The comparison of the determined values with data of other biofuels (2-methylfuran, 2-methylterahydrofuran) showed a higher toxicity of the mixture. DnBE show a higher toxicity than D. magna as the tested mixture. For the comprehensive ecotoxicological assessment of the mixture and three compounds as single substances further tests are required. Especially regarding to the possible impairment of the hatch rate of D. rerio. Preventing of slippage can cause the larvae to be no longer viable, resulting in a misinterpretation of the detected LC50. In future, further ecotoxicologically relevant endpoints should be investigated. This was performed as part of the project "Tailor made fuels from biomass" funded by the Excellence Initiative by the German federal and state governments to promote science and research at German universities.
inherent to the use of the insecticide abamectin and the fungicide difenoconazole, the main objective of this research was to evaluate the effect generated in Danio rerio embryos exposed to pesticide mixtures and evaluate the effects produced by the interaction of these compounds. For this, Danio rerio embryos were exposed for 96h to the binary mixtures of abamectin and difenoconazole following the recommendations of OECD TG 236. The concentrations used were 0.5; 1.1; 2.5; 5.3 and 11.7 mg L$^{-1}$ of abamectin and 0.2; 0.5; 1.0; 2.5 and 5.0 mg L$^{-1}$ of difenoconazole. The factorial design was used to cover all possible concentrations, and in total 35 treatments plus the control were performed. The exposures were performed in 50 mm Petri dishes using three plates per treatment. In each plate containing 15 mL of solution were arranged 5 eggs totaling an n = 15. Survival data were recorded every 24 hours and the results were analyzed in the Minitab software. The results showed that the binary mixtures of abamectin and difenoconazole promote in Danio rerio embryos the antagonistic effect in the lower concentrations, but in higher concentrations the produced effect is synergic. This means that, at lower concentrations the interaction of abamectin + difenoconazole seems to decrease the toxicity of pesticides to Danio rerio embryos, but the toxicity of the compounds is potentiated at higher concentrations of the mixtures. Similar results were obtained in other studies with compounds each exposed to mixtures of these same compounds, but complementary studies are necessary to better understand the toxicokinetic of these pesticides mixtures.

**TH302**

Cocktail-effect of persistent organic pollutants on selected bioreporter-systems and zebralife embryos

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There is an increasing number of chemicals including pharmaceuticals and industrial pollutants that are released into the aquatic environment, leading to the exposure of fish and other aquatic organisms. Moreover, at the present time environmental risk assessment is mainly based on chemical analysis, only. However, “compound-by-compound” based assessments seriously run the risk of underestimating the risk of chemicals as the true exposure scenario for humans and wildlife is known to be far more complex. Under regular environmental conditions organisms can be exposed to multiple chemicals associated with different risks and specific effects, e.g. teratogenicity, immune toxicity and suppression, genotoxicity, and endocrine disruption. Moreover, it has been repeatedly demonstrated that pollutants and the underlying toxic responses may interact and generate effects that are different from the toxicity of the individual chemicals. Thus, understanding the effects of simultaneous pollutants generally referred to as “cocktail effects”, represent one of today’s greatest challenges in environmental but also in human toxicology. The aim of the present study is to investigate embryotoxic and teratogenic, but also mechanism-specific effects using zebralife embryos. They will be exposed to selected priority pollutants and their mixtures (e.g. polychlorinated biphenyls, heavy metals, polycyclic aromatic compound). These chemicals represent highly relevant chemicals which can be found in great levels in the environment. First results indicate that beside biological interactions heavy metals may alter the toxicity of organic pollutants. This study is part of the EnForce project (https://www.oru.se/enforce), which aims at the development of an effect-based risk assessment in cooperation with different stakeholders and several industrial partners.

**Emerging technologies and related raw materials requirements scenarios: the role of life cycle thinking (P)**

**TH304**

Environmental impact assessment of carbon fibers reinforced composites: pyrolysis process


The end-of-life management of carbon fibers reinforced composites (CFRCs) has been investigated by comparing the environmental sustainability of Curti S.p.A. company’s pyrolysis process with waste-to-energy (WtE) and landfill disposal. The determination of environmental loads was carried out through the Life Cycle Assessment (LCA) methodology, modeling and analyzing each scenario throughout a cradle-to-grave and cradle-to-container database. CFRCs are highly engineered materials, with high calorific power and excellent mechanical properties. From their recovery, it is possible to obtain a secondary raw material that can be used in application requiring lower performance than originals, or the recovery of thermal/electrical energy. The market still offers few CFRC recovery technologies. Therefore the most developed ones have been chosen to compare with landfill disposal, even though nowadays it would be avoided, for waste with a LHV $>13$ MJ kg$^{-1}$. The pyrolysis process involves a first pyrolysis and a subsequent gasification of the waste within the same reactor. This system allows the quantitative recovery of carbon fibers (CF) contained in the initial composite and generates hot gaseous compounds that are burnt and released into the atmosphere. Since the plant is at a pilot scale, a heat recovery system has not been designed yet for the combustion of fumes. Considering the impact assessment results, pyrolysis has proved to be the most sustainable treatment due to the quantitative recovery of carbon fibers, which avoids the consumption of material and energy deriving from the production of virgin carbon fibers. The worst scenario is WtE, mostly due to the emission of CO2, NOx and SOx generated by emissions in air and water. The impact of landfill disposal is intermediate, due to the good stability of CFRCs: having a slow degradation, their disposal in landfills does not cause a high impact, except for land occupation. The LCA study made it possible to carry out a preliminary assessment supporting the pyrolysis pilot plant design, to identify critical aspects and strengths of each scenario. [1] Legislative Decree n°36 of 13 January 2003; Implementation of Directive 1999/31/EC on landfill of waste, Official Journal of the Italian Republic, 2003. [2] Legislative Decree n°205 of 3 December 2010; provisions implementing Directive 2008/98/EC of the European Parliament and of the Council of 19 November 2008 on waste, Official Journal, 2010.

**TH305**

Critical raw materials in a new building integrated photovoltaic system

D. Garrain, I. Herrera, Y. Lechón, CIEMAT / Energy Dept

**Energy Systems Analysis Unit REELCOOP, an EU-FF7 funded project which stands for RENEWable ELectricity COOPeration (www.reelcoop.com), aims to develop and test novel prototypes of electricity generation technologies. One of the prototypes is a solar photovoltaic (PV) ventilated façade (6.6 kW) and involves the development of e-Si solar cells, as well as the study of the ventilation effect in PV façades. PV solar panels have particular metals or rare earths that are potentially included in the category of 'critical raw materials (CRMs)'. This work aims to identify the potential CRMs in this prototype and to define several ways to improve the sustainability from a life-cycle approach, including aspects like substitution or recycling of these materials.

**TH306**

Environmental sustainability assessment of a biological Active Pharmaceutical Ingredient: A resource based Life Cycle Assessment

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Most Life Cycle Assessments (LCAs) performed in the pharmaceutical sector have been done on chemically synthesized drugs, leaving biopharmaceuticals aside. The fact that sustainability assessments of biopharmaceutical products and technologies have been rarely done is probably a consequence of the major challenge that building a robust Life Cycle Inventory (LCI) represents for the upstream and downstream processes of biologics. However, the low number of LCAs performed on biopharmaceuticals does not reflect the current reality of the pharmaceutical sector, as its market is rapidly growing at an annual rate of around 9%. Therefore, this study focuses in performing a resource based LCA to measure the environmental impact of a biological Active Pharmaceutical Ingredient (API). A resource based methodology is applied for addressing the task that additionally focuses on supply and efficient use of resources. The API investigated is infliximab, a monoclonal antibody that treats autoimmune diseases. An Exergetic Life Cycle Assessment (ELCA) was conducted, using the Cumulative Exergy Extracted from the Natural Environment (CEEENET) method. First results show that the unit operations with the highest impact are: i) The first chromatographic process for purification (Direct Product Capture), since it requires the highest quantity of buffers which are produced using chemicals as well as complex organic compounds such as amino acids. ii) Fermentation, as similar complex components are required for its medium, which are also produced through biotechnological processes. Furthermore, fermentation is the process that takes the longest (several days), leveraging the Heating Ventilation and Air Conditioning (HVAC) system to achieve the clean room conditions needed for the production of biologics. HVAC has shown to be the utility with the highest impact, consuming a significant percentage of the total electricity used in the plant. Performing an LCA on a biologic mainly using primary data has been proven to be a possible task. However, challenges such as the data unavailability of biotechnologies used to produce the nutrients needed throughout the process, as well as the further integration of these technologies into databases should be addressed.

**TH307**

LCA methodology: a case study of the industrial production of terephthalic acid from renewable sources

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**Innovation Lab S.r.l.; E. Neri, Alma Mater Studiorum - University of Bologna; F.**
The scope of the present study is to investigate the environmental sustainability of different routes of terephthalic acid (TA) production, comparing the results achieved for both the traditional way with those of three bio-based routes. The aim of the study is to identify which of the selected pathways has the lowest environmental load. Below the four routes selected are briefly described: Traditional way: p-xylene is obtained from catalytic reforming of crude oil as part of extracted BTX (benzene, toluene and xylene isomers); GEVO’s process: isobutanol from the fermentation of biomass is converted into hydrocarbons, iso-octene and p-xylene; Friedel-Crafts for HMF and ethylene: it involves the production of HMF (S-hydroxymethylfurfural) from starch, its conversion to DMF and the Diels–Alder reaction with bio-ethylene to obtain p-xylene; Alternative pathway from p-cymene: it consists in the oxidation, using O₂ in the presence of a catalyst, of p-cymene (derived from orange peels) to obtain TA. The first two routes are already set at industrial level; the others are still under development. Therefore, in order to estimate the environmental performance of the scenarios, a simulation of the sustainable processes was carried out using ChemSolv software. The first three syntheses are based on the production of p-xylene, subsequently oxidized to TA through the Amoco process. On the contrary, the last route is proposed as an alternative way, since it uses p-cymene as a precursor of TA. In order to perform the comparison, a LCA (Life Cycle Assessment) methodology has been used as a scientific tool of energy capacity, residence of a pictorial report in each scenario. LCA was able to evaluate the environmental performance integrated with the entire life cycle of a product, in our case 1 ton of terephthalic acid. The model was validated by the use of SimaPro software and the results indicate that bio-based processes are sustainable only if they use organic residues as raw materials. Both analysis methods used, CED and ReCiPe, have shown that only the alternative route from orange peels has lower environmental impacts than the traditional way from crude oil. For the other processes, the cultivation and transformation of biomass present an environmental weight that does not justify their use as starting materials for p-xylene, and then TA, production. Thanks to this work it was also possible to make some considerations about energy requirements and their weight in the life cycle of the processes.

**TH308**

**Environmental assessment of vanadium redox flow batteries**

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The rising production of electric energy from renewable sources requires electrical energy storage systems to compensate for the fluctuations in energy generation. The vanadium redox flow battery (VFB) is a suitable technology for stationary energy storage on a broad scale. Due to the independent scalability of system power and energy capacity, residential and industrial systems can be considered, in order to identify the key parameters for a deliberate green process. Additionally, the cultivation and transformation of biomass present an environmental weight that does not justify their use as starting materials for p-xylene, and then TA, production. Thanks to this work it was also possible to make some considerations about energy requirements and their weight in the life cycle of the processes.

**TH309**

**Towards the Life Cycle Assessment of engineered nanoparticles production: a comparison between batch and continuous flow synthesis**

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The aim of this study is to provide an assessment of continuous micro/milli-flow technologies applied to Engineered Nanoparticles (ENPs) production, benchmarking them against the conventional batch productions. As the quantity of ENPs produced and their applications are steeply growing, an increasing attention is being paid to the quality of the product and the efficiency of the synthesis. Continuous micro/milli-flow (CF) synthesis is considered to be the natural evolution of the currently most diffused conventional batch synthesis, as it intensifies the production in terms of productivity, energy and chemicals use, product quality and functionalisation. Given the importance and potentiality of the transition, an evaluation of these new technologies is needed. Life Cycle Assessment is the chosen methodology for addressing this objective, as it looks deep into the sustainability, efficiency and environmental impact of the system considered, in order to identify the key parameters for a deliberate green process design. The application of the LCA methodology to ENPs production presents many challenges: to date, few LCA studies within the manufacturing process of ENPs have been carried out. In this work, a wide selection of ENPs production processes have been evaluated and compared, using CED and ReCiPe. The CF syntheses are evaluated on a lab scale, performing a hot-spot analysis and benchmarking them against the equivalent batch syntheses. The output of the assessment permits valuable considerations on the best equipment materials, solvents, stabilizers, type of heating and mixing for maximising the efficiency of the process, even at the very early stages of its development. The results obtained highlight a general favourable tendency toward the CF system as a greener and more efficient way of ENPs synthesis than the correspondent batch production.
High-operating-temperature thermal storage materials for TES increasing up to 3 times the thermal capacity. All these solutions are being assessed through a comprehensive LCA, considering the entire life cycle of materials and components, from raw material extraction until the end-of-life. A comparative analysis is being prepared between baseline scenario (with reference materials) and the scenario with the IN-POWER innovative materials. Along the project different candidate materials and approaches are being assessed and a decision making process looking for high performance materials but environmentally friendly. Some improvements are being made such as: use of aluminum instead of silver in the mirror reflective coating, to meet the European requirements for use of non-critical materials; the reduction of materials weight; and the increase in materials robustness. The expected results are to: Obtain a complete environmental profile of IN-POWER CSP architecture. Calculate the environmental impacts associated to: new polymeric materials for mirrors; high absorber coating; high thermal storage capacity materials; polymeric composite for CSP structure. Evaluate the benefits of IN-POWER materials compared with reference materials. Evaluate the benefits of IN-POWER CSP architecture compared with current mature Parabolic Trough Collector technology.

TH312 Environmental impact and social influence of an Advanced Adiabatic Compressed Air Energy Storage (AA-CAES) located in Eisenerz, Austria. The case of RICAS2020 PROJECT. A. Claret, Leitat Technological Center / Sustainability Division; G. Ferrer, Leitat Technological Center / Quantitat Ge on / RD Safety / Social impact and benefits of AA-CAES; M. Riera, LEITAT / Sustainability Division; S. Vazquez, ACONDICIONAMIENTO TARRASENSE (LEITAT TECHNOLOGICAL CENTER) / Sustainability Division

European society has a highly dependency on electric power. In 2009 European Union fixed that at least 20% of EU gross final energy consumption have to come from renewable energy sources until 2020. The increasing use of renewable energy sources to produce energy has generated a worldwide challenge: (i) a first stage of improved enhanced biogas flare removal with upgrader recovery and (ii) a second stage of autotrophic biological nitrogen removal in two-steps. A life cycle assessment will determine the environmental impacts and benefits of this upgraded system with respect to the conventional setup. Policy tradeoffs will also be assessed. This wastewater treatment plant originally benefited from cogeneration, but eventually decided to use biogas flare systems due to the taxes on self-supply of electricity imposed by the Royal Decree 900/2015 approved by the Spanish Parliament. As a result, we need to determine if applying technologies that generate more biogas will result in larger impacts than the current scenario if the biogas cannot be used for cogeneration.

TH314 Anticipatory life cycle assessment of sol-gel derived anti-reflective coating for greenhouse glass N. Tsay, CML Leiden University / CML; J. Quist, Delft University of Technology / Technology, Policy, and Management; A. Wypkema, M. Mourad, TNO / Materials Solutions; V. Prado, CML Leiden University

Technological innovations can provide sustainable development but at the same time it can cause unfavorable consequences to environment and society. Environmental assessment of the technologies is usually performed when they have already been launched in the market with a low possibility to transform their development towards better environmental performance. Anticipatory Life Cycle Assessment (LCA) has been evolved to assess the environmental impacts of the technologies before they are introduced in the market. This work is focused on the weighted assessment of two different greenhouse glass coatings that are used for glasshouse applications: the anti-reflective coating and the conventional coating. The study on the anti-reflective coating is based on applied scientific research. The conventional coating is based on a scientific approach. As a result, more yield of crop could be obtained under coated glass. The Netherlands Organization for Applied Scientific Research (TNO) is developing a novel anti-reflective coating for greenhouses which is expected to have higher light transmittance than the conventional coatings by 2.5%. In this study, three reference coatings in addition to the novel coating were assessed. The functional unit was the mass of tomatoes obtained under a certain area of greenhouse glass coated with uncoated/coated glass during 30 years. The novel coating is being synthesized in the laboratory scale, and thus, in LCA, What-if scenarios were used to scale-up the coating system to pilot and industrial scales. The laboratory parameters, e.g. the amount of electricity used to produce the coating and the solution volumes, were optimized using literature and expert consultation. The comparative analysis methodology is an enhanced version of the conventional LCA, and this could be due to the simplicity of the coating method applied by TNO. Also, it was revealed that the coatings do not bring significant environmental benefits rather they bring economic benefits in terms of increased yield of tomatoes. Finally, the sensitivity analysis showed that electricity used for the production of glass has higher impacts than transmittance or degradation time of the coatings.

TH315 Combined process simulation with Life Cycle Assessment method in polyurethane rigid foam production A. Bordignon, M. Fermeglia, Università di trieste; A. Bortolazzi, S. Rondinini, C. Locatelli, A. Vertova, Università di Milano

Process simulation is a computational technique used in several sectors of process engineering. It is very useful at design stage, for defining the feasibility of a new process, as well as in the process optimization stage, where the optimum value of the production is pursued and, last but not least, in the process analysis stage for understanding the potential improvement of an existing process. It consists in the coupling of thermodynamic simulations (using an explorative approach). Here, we show the application of Anticipatory LCA in the assessment of the anti-reflective greenhouse glass coatings. The light intensity is essential for the crop growth in the greenhouses. A portion of the light is lost when it reaches the greenhouse glass surface due to the glass reflection. The anti-reflective glass coatings can be applied on the surface of the greenhouse glass to allow more sunlight to enter the greenhouses. As a result, more yield of crop could be obtained under coated glass. The Netherlands Organization for Applied Scientific Research (TNO) is developing a novel anti-reflective coating for greenhouses which is expected to have higher light transmittance than the conventional coatings by 2.5%. In this study, three reference coatings in addition to the novel coating were assessed. The functional unit was the mass of tomatoes obtained under a certain area of greenhouse glass coated with uncoated/coated glass during 30 years. The novel coating is being synthesized in the laboratory scale, and thus, in LCA, What-if scenarios were used to scale-up the coating system to pilot and industrial scales. The laboratory parameters, e.g. the amount of electricity used to produce the coating and the solution volumes, were optimized using literature and expert consultation. The comparative analysis methodology is an enhanced version of the conventional LCA, and this could be due to the simplicity of the coating method applied by TNO. Also, it was revealed that the coatings do not bring significant environmental benefits rather they bring economic benefits in terms of increased yield of tomatoes. Finally, the sensitivity analysis showed that electricity used for the production of glass has higher impacts than transmittance or degradation time of the coatings.

TH316 Life Cycle Assessment of CO2-based Methanol Production using Captured CO2 from Fossil Fuel Power Plants C. Lee, University College London / Department of Chemical Engineering; R.
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As a way to address the climate-related CO2 emissions from fossil fuel power plants, photocatalytic methanol production using a novel form of CO2 conversion process has been investigated within the research project “Low Carbon Fuel”. The primary goal of this study is to evaluate the environmental performance of photocatalytic CO2 conversion in comparison with conventional fossil-based technologies for power generation and methanol production. Life cycle assessment (LCA) is used to determine and compare the environmental performance of the methanol production systems. In the LCA study, cradle to gate system boundaries are used because the downstream processes and properties of methanol are similar for CO2-based and fossil-based systems. Since the main environmental motivations for photon utilisation are reducing CO2 emissions and establishing an alternative carbon source, this study compares the CO2-based and fossil-based methanol production systems with respect to global warming and fossil resource depletion. The CO2-based methanol production system consists of the following three stages: CO2 source including CO2 capture, electricity compensation, and CO2 utilisation for methanol production. The fossil-based methanol production system serves as benchmark and is divided into electricity generation and fossil-based methanol production. The main functions of the CO2-based and fossil-based systems are production of methanol, and supply of electricity to the UK electricity grid. To quantify the main functions, we choose 14.3 MJ methanol (655 g) as reference for the function ‘methanol production’. The second function ‘electricity supply’ can be quantified through the amount of CO2 that is captured to produce equivalent amount of methanol. CO2-based and fossil-based methanol production processes are analysed and compared based on life cycle assessment. Our analysis reveals that CO2-based methanol production system using photocatalytic CO2 conversion is not always mandatory to achieve CO2-based system with lower environmental impacts than the fossil-based system. However, CO2-based methanol production has the potential to reduce impacts for global warming and fossil depletion if the environmental performance of the intermediate steps and steps are improved, compared to the corresponding fossil-based route. Furthermore, additional environmental benefits can be obtained from environmentally friendly hydrogen production from photocatalytic water splitting process.

Advancing the Adverse Outcome Pathway Framework - An International Horizon Scanning Approach (P)

TH317

Linking failed swim bladder inflation of larval Japanese medaka (Oryzias latipes) after embryonic exposure to 17α-ethinylestradiol, levonorgestrel and dienofuran, to disrupted β-fetuin/Wnt signalling

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Embryotoxicity testing is a high throughput alternative to using the whole fish model. Previous studies in our laboratory have demonstrated that embryonic exposure to pharmaceutical compounds capable of disrupting the endocrine system such as 17α-ethinylestradiol, levonorgestrel, and dienofuran, both alone and in mixtures, can impair swim bladder inflation of Japanese medaka (Oryzias latipes). Failure of swim bladder inflation can have serious long-term effects on fish population and is linked with increased mortality. Embryonic exposure to xenobiotic compounds are able to cause swim bladder inflation is not fully understood; however, it is possible that compounds are able to cause their effects through a disruption of embryonic cell signalling pathways. The canonical Wnt pathway plays a crucial role in fish swim bladder development, and the disruption of Wnt signalling during swim bladder organogenesis could lead to improper swim bladder formation. The effects of two Wnt modulators IWR-1 and lithium exposed from 36-101 hours post fertilization on gene expression of medaka embryos was determined. The effects of embryonic exposure to the three pharmaceutical compounds on whole embryo gene expression were then established and compared. It was measured that these pharmaceutical compounds significantly inhibited the expression of genes related to the formation of the three layers of the swim bladder (epithelial, mesenchyme, and outer mesothelium). Both levonorgestrel and 17α-ethinylestradiol also significantly downregulated the expression oflef1 and tcfl7 (β-catenin/Wnt transcription factors), but not the expression of the Wnt ligand wnt5b. Thus demonstrating that these compounds may be altering swim bladder inflation through a disruption of β-catenin/Wnt signalling during early embryo development.

TH318

Linking mode of action of the model respiratory and photosynthesis uncoupler 3,5-dichlorophenol to adverse outcomes in Lemma minor

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Lemma minor is an aquatic plant commonly used in laboratory phytoxicity testing due to its rapid reproduction capacity, resource-effective exposure format and central function in the aquatic ecosystem. Several standard methods have already been adopted by international standardisation organisations using this species as an ecological model. Although being highly useful for regulatory purposes focusing on traditional adverse endpoints, these tests provide limited information about the toxic mechanisms and modes of action (MoA) and rarely address complex environmental issues such as exposure to multiple stressors. The present study aimed to use selected functional assays in L. minor after exposure to 3,5-dichlorophenol (3,5-DCP) as a model to characterize the toxic mechanisms causing growth inhibition and lethality in primary producers. The results demonstrated that 3,5-DCP caused concentration-dependent effects in chloroplast and mitochondria. Endpoints such as uncoupling of oxidative phosphorylation (OXPHOS), chlorophylls content, reproduction rate and frond size are more sensitive to 3,5-DCP compared to other responses as well as reactive oxygen species (ROS) formation, lipid peroxidation (LPO) and impairment of photosynthesis efficiency. Principal component analysis (PCA) indicated that suppression of photosystem II (PS II) efficiency, electron transport rate (ETR), ROS production and LPO, pigments content and growth were strongly correlated while inhibition of oxidative phosphorylation (OXPHOS) which was more closely correlated with growth parameters. A set of conceptual Adverse Outcome Pathways (AOPs) were developed by using Bayesian network model to decipher the causal relation well studied in vitro endpoints, especially mammalian species. However, for other ecologically relevant environmental species, such as aquatic invertebrates, the knowledge is still limited. The present study was therefore conducted to: develop AOPs for ROS-mediated reproductive effects in aquatic invertebrates based on existing data from the literature; assess the weight of evidence (WoE) of the AOPs based on a combination of literature survey and in silico predictions; evaluate the knowledge gaps in the AOPs by using the forecaster species Daphnia magna. An extensive literature survey to collect existing knowledge on ROS-mediated reproductive effects in aquatic invertebrates, and metals, ionizing and non-ionizing radiations as prototypical stressors. A network of conceptual AOP was assembled first and a linear AOP with the strongest data support was then selected prior to WoE assessment using the Evolved Bradford Hill considerations. The laboratory evaluations of the AOPs were performed using UVB and gamma radiations as prototypical oxidative stressors. Daphnia magna was used as the model species. The laboratory studies clearly showed that both UVB and gamma radiation caused dose-dependent reduction in reproduction after the exposure. The reproductive effects were correlated with excessive ROS production, lipid peroxidation, DNA damage, apoptosis, mitochondrial respiratory chain, protein and lipid storage, and abnormal ovary structures and oogenesis in D. magna, thus verifying several KEs in the conceptual AOPs. This study has for the first time systematically linked excessive ROS production to reproductive effects in aquatic invertebrates using the AOP concept, thus providing mechanistic knowledge for future hazard and risk assessment of oxidative stressor-mediated adverse effects in ecologically relevant species.

TH320

Development of an Adverse Outcome Pathway for cardiotoxicity mediated by the blockade of L-type calcium channels

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A diverse set of chemical compounds, including some pharmaceuticals and...
insecticides, have the potential to perturbate the functionality of calcium channels. Among the different types of calcium channels, the L-type calcium channel (LTCC) is responsible for the excitation-contraction coupling of skeletal, smooth, and cardiac muscle. Chemicals that unintentionally block this channel in cardiac cells may impair heart function and health, leading to various cardiac pathologies and predisposing individuals to heart failure. Advancing our understanding of the mechanisms underlying these adverse effects is of paramount importance if we want to develop effective strategies able to accurately predict the cardiac risk posed by chronic exposure to those chemicals. In this presentation, we describe the development of an Adverse Outcome Pathway (AOP) that outlines the series of causally related key events triggered by the blockade of LTCC, and that can ultimately lead to cardiac adverse effects. We discuss the integration of in silico, in vitro, and in vivo evidence to support the AOP development, as well as the application of computational and network biology approaches that may accelerate the identification of relevant key events. Considering the multifaceted role of LTCC in different components of the cardiovascular system other than the heart, we also discuss the importance of applying AOP network considerations to guide a reliable and fit-for-purpose AOP development. This AOP will represent a valuable knowledge base able to guide the identification of key events that are highly predictive of in vivo toxicity, and that can be measured in vitro without relying on animal testing. The knowledge base will also be used as platform to drive future development projects aimed at incorporating additional layers of complexity in the model, and at driving the transition towards a fully quantitative AOP able to effectively support regulatory decision-making and risk assessment.

**TH321**

**Quantification of AOP by Bayesian network modelling: linking 3,5-DCP exposure to adverse outcomes in Lemma minor**

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APs have gained international recognition as a systematic approach for capturing existing toxicological knowledge to transparently link mechanistic data to toxicity endpoints. Nevertheless, most AOPs are qualitative and not directly suitable for quantitative risk assessment. Quantitative AOPs (qAOP) should define the relationships underlying transition from one KE to the next sufficiently well to allow quantitative prediction of the probability or severity of the AO occurring for a given activation of the MIE. We have started developing a Bayesian Network (BN) model to quantify a recently proposed AOP, which links the mode of action of the model respiratory and photosynthesis uncoupler 3,5-dichlorophenol (DCP) to adverse outcomes in the aquatic plant Lemma minor. The BN model is based on data from a laboratory experiment exposing L. minor to DCP in 8 concentrations with 3 replicates. The measured response variables include OXPHOS (oxidative phosphorylation), ROS (reactive oxygen species), ETR (electron transfer rate), Fv/Fm (maximum quantum yield of photosystem II), LPO (lipid peroxidation) and number of fronds (leaves). The proposed AOPs a network consisting of three chains with the same chemical stressor (DCP) and AO (fronds number). All AOP components are defined in the BN as nodes with discrete states. Each node is quantified by a probability distribution across these states. The causal links (Key Events Relationships) are quantified as conditional probability tables (CPTs), which determine the probability distribution of a child node conditionally on the probability distribution of the parent node(s). The CPTs are calculated directly from the data in this BN version. The BN was run by changing DCP concentration and inspecting the changes in all subsequent nodes. Qualitatively, the model predictions were as expected: increasing the DCP concentration caused reduced OXPHOS, reduced ETR and reduced fronds number. For example, when DCP was increased from 1 to 2 mg/L, the probability of fronds number being in the lowest (worst) state increased from 6% to 30%. Fine-tuning of the intervals of some nodes is needed to make the BN more responsive. Moreover, we will use statistical approaches to obtain more credible CPTs, such as estimation of dose-response curves with uncertainty. Other planned developments include linking the AO to an endpoint with regulatory relevance, and linking the chemical stressor to an Aggregate Exposure Pathway.

**TH322**

**Development of Quantitative Adverse Outcome Pathway (AOP) of Pulmonary Fibrosis with Effectcopedia**

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Pulmonary fibrosis (PF) is a chronic and progressive lung disease where the scars are formed in the lung tissues and the air sacs in the lungs (alveoli) becomes stiff leading to serious breathing problems. Several substances are identified as inducer of PF, but high cost of inhalation toxicity studies refrain to conduct systematic studies of all those substances. Hence, the regulations of these substances become obscure. To solve this problem, Adverse Outcome Pathway (AOP) concept has been emerged. AOP is a framework that organizes existing knowledge about linkage between molecular-level perturbation and an adverse outcome. To facilitate the development of AOP, OECD launches AOP knowledge-base (KB). In recent years, the application of quantitative AOP (qAOP) which provide dose-response and time-course prediction, has been gaining much more attention in regulatory decision-making field. To develop the AOP of pulmonary fibrosis, in one hand, we made preliminary AOP from literatures, which constitutes the PPARγ interaction as Molecular Initiation Event (MIE), Collagen activation, Inflammation and EMT-Fibrosis activation as Key Events (KEs), and Cytotoxicity/Apoptosis and Fibrosis as Adverse Outcome (AO). On the other hand to make qAOP, we conducted cytotoxicity and apoptosis test using human bronchial epithelium cell (Beas2B). Beas2B cell was exposed to CMT/MIT (a biodegradable substance which possess potential risk to respiratory systems) at various doses from 0 mg/L to 2 mg/L for various time for 1 to 72 hr. Cytotoxicity and apoptosis was analyzed using various available assays at mid to high through put condition. While, quantitative analysis of the KEs was performed using Effectopedia platform of AOP-KB. For further study, we are planning to do various dose- and time response test (using qPCR and ELISA) for each potential KEs, so that we can integrate data for building qAOP model with the network between MIE-KEs-AO. Acknowledgement: This work was supported by a grant from the Korean Ministry of Environment through ‘Environmental Health R&D Program’ (2017001370001).

**TH323**

**Exploring Potential of Knowledge Databases for Adverse Outcome Pathway Discovery**

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Adverse outcome pathways (AOPs) have potential to support and enhance the use of mechanistic data in regulatory decision-making. AOPs organize existing knowledge about relationships (ideally causal ones) between initial chemical-biological interactions (molecular initiating events; MIEs), intermediary key events (KEs), and adverse outcomes (AOs) relevant to risk assessment. Efficient ways of AOP development and weight of evidence assembly are lacking. This study evaluated potential of the existing knowledge databases (Unified Medical Language System - UMLS, and National Library of Medicine – NLM) for AOP discovery and development. UMLS contains more than 68-million relationships among more than 3-million unique biomedical concepts (or terms). The NLM literature database contains more than 100-billion relationships among similar 3-million biomedical concepts extracted from the abstracts of more than 16 million biomedical journal papers. First, AOP network was downloaded and parsed from AOP Wiki (https://aopwiki.org/). We found that there are 3,084 relationships among stressors, MIEs (main initial events), KEs (key events), AOs (adverse outcome), stressor-chemicals, and stressor-events. High performance graphic processing unit (GPU) was used to determine which of 3,084 relationships can be found in hundred million of relationships in UMLS and NLM databases. 610 (20%) relationships were found in the UMLS database. About 1,837 (60%) relationships were found in the abstracts of 16 million biomedical papers on NLM. When combining our searches over both the UMLS and NLM databases, 1,983 (64%) relationships from AOP wiki were found; relationships in some sub-categories such as stressor-chemicals had much higher hit ratio - 78%. These findings indicate that AOP discovery system that uses UMLS and NLM to predict new probable AOP relationships (that can connect to objects in the AOP Wiki) could substantially accelerate AOP development and contribute to weight of evidence analyses. The confidence of the predicted relationships could be calculated based on frequency of the relationships, whereas the quality of the predicted relationships could be further improved by training deep learning models with knowledge curated in databases such as The Comparative Toxicogenomics Database, ECOTOX and iCSS ToxCast Dashboard.
Fish model species in human and environmental toxicology (PC)

MOPC01 Fish caging experiment as a tool for in situ assessment of neurotoxic effects of untreated wastewaters

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In the framework of FP7 project Solutions, the city of Novi Sad has been selected as a pollution hot spot of the River Danube, mainly due to the direct discharge of untreated sewage into the river. Significant biological effects of untreated wastewater were previously observed through an in vitro study. To study neurotoxic effects more in details and in realistic ecological context, we conducted fish caging experiment. Common carp (Cyprinus carpio (L.), Cyprinidae) has been selected for the experiment, as one of the most common species in the Middle Danube, genetically well described and economically important. Ten fish per cage were exposed in cages for nine days at three sites on the Danube River: upstream from sewage discharge – reference site, 230 m and 7 km downstream from the discharge and at one site on Sava River (downstream from industrial wastewater discharge near the city of Sabac). After the exposure period, brains were isolated and prepared for gene expression and enzyme activity analyses. The expression of genes encoding for five proteins was studied: tachykinin 3a and tachykinin 3b (involved in neuroendocrine regulation of reproduction), GABA a1 receptor (for the inhibitory neurotransmitter GABA and various drugs), synaptotagmin 10 (integral membrane protein of synaptic vesicles with a role in exocytosis) and myelin basic protein (responsible for myelination of axons and neuroprotection). The activity of acetycholine esterase, enzyme that terminates action potential transmission in chemical synapses of cholinergetic type, was also examined. A trend of slight upregulation for the expression of the genes encoding for tachykinin 3a and tachykinin 3b, GABA a1 receptor and synaptotagmin 10 was observed for all three studied sites when compared to the reference site. The expression of the gene encoding for myelin basic protein was similar at reference site and 230 m downstream from the sewage discharge, but this gene expression was significantly downregulated downstream from the industrial wastewater discharges. Based on this result, myelin basic protein might be potential selective biomarker which can be used to differentiate the effects of these two types of chemical pressure. No significant difference was observed in the activity of the acetylcholine esterase between studied sites. The study is part of the SOLUTIONS project, funded by the EU FP 7 (FP7-ENV-2013-two-stage Collaborative project) under grant agreement number 603437.

MOPC02 Toxicity analysis of treated sugar cane vinasse by integrated systems using gills of Oreochromis niloticus as model

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The algal fermentation of sugar cane (Saccharum sp.) results in a by-product known as vinasse. This by-product is used as fertilizer because of its richness in organic matter, and also because it promotes improvement in soil fertility, favoring the availability of some elements for the plants. However, the amount of vinasse used in the fertilization should not overcome the ion retention capacity of the soil, since the dosages should be directed to the specific characteristics of each soil. When used in unbalanced proportions can impair to the soils and the plants, in addition to being able to reach water resources. Considering studies that prove the toxicity of vinasse in nature, the use of treatment systems has become quite interesting. The integration of systems such as natural attenuation, filtration and phytoremediation increase the effectiveness of the treatment, since they are highly effective biogeochemical systems to treat waste water from different sources. Aquatic macrophytes, which not only accumulate pollutants directly in their tissues but also act as catalysts for purification reactions that usually, occur in the rhizosphere of plants, are part of the alternative treatment for vinasse. Fish are excellent experimental models for aquatic toxicity studies because they warn of the potential danger of chemicals reaching water resources. Therefore, this study aimed to verify the efficacy of sugar cane vinasse treatment in reducing its toxic potential by histological and histochemical tests on tilapia gills. The animals were submitted to two different dilutions of the treated vinasse for 96 hours; after this period the gills were removed and submitted to standard histological routine. Morphological analyses of the gills revealed that the cellular pattern described for the species was not altered and histochemical tests showed a decrease on number of mucous cells, thus attesting to the decrease on toxicity of the treated vinasse. Thus, it can be inferred that integrated treatment systems were effective in reducing the polluting potential of vinasse, since the animals did not present histological changes.

MOPC03 Assessing toxic effects in the fish Violet Goby (Gobioides broussonetii - Gobiidae) from one of the most productive estuaries in Brazil

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The Estuarine-Lagoon Cananéia (São Paulo, Southeast Brazil) is among the most productive areas in the South Atlantic. The Ribeira de Iguape River (RIR) is the major freshwater contributor of the estuary. It carries different classes of contaminants from former mining activities, agricultural areas and urban centers through an artificial channel. The disordered human occupation, presence of boats and the disposal of waste and sewage are also sources of pollution throughout this system. The Violet Goby (Gobioides broussonetii - Gobiidae) is a demersal fish of a social and economic importance to traditional fisheries. Over the last decade the regional disappearance of this fish species has been reported, including events of high mortality. The contamination by the RIR has been referred as one of the causes of the decline of that population. Therefore, this study aimed to observe possible toxic effects in G. broussonetii in the studied area. Fishes were sampled near Cananéia, Subaúma and Iguape in winter (2016) and summer (2017). The animals were anesthetized, euthanized and the blood, brain, muscle, liver and kidney were collected for the biochemical and genetic biomarkers analysis. The muscle AChE activities showed similar results among the points and seasons, while cerebral AChE were lower in Subaúma in winter. Hepatic and renal GPs and GST activities, GSH concentrations and LPO damage in liver were similar. However, it was observed genotoxicity (micronuclei, DNA breaks, and sister chromatid exchanges) in liver and blood in summer. In winter, nuclear morphology alterations were identified in erythrocytes more frequently in fishes of Cananéia. The results suggest that contaminants such as metals and HPAs previously reported in the sediments may have been stressing this species. The marked seasonality of the region, which consequently influences the temperature, the rainfall regime and the bioavailability of contaminants may interfere in the obtained results. Nevertheless, an antihypoxic activity was observed near to Cananéia and Subaúma points. It suggests an influence of the local hydrodynamics by dragging the contaminants of the main sources (RIR and Cananéia city) to these areas once lower impacts were seen in Iguape. This last point is located above the artificial channel in an area of low hydrodynamic. Water and sediment chemical analyses are being performed in the studied points in order to support a better understanding of these responses.

MOPC04 Does ozonation of the Aachen Soers WWTP improve the water quality in the field? Caging experiments with juvenile rainbow trout and various biomarker approaches

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The Aachen Soers WWTP is a treatise aimed to achieve a good ecological and chemical state of all surface waters until 2015. However, the good biological state is only reached by around 20 % of the German surface water bodies. One reason might be the release of a variety of anthropogenic contaminants by waste water treatment plants (WWTPs) into these water bodies. Additionally, these substances are not sufficiently eliminated via conventional waste water cleaning processes. Nevertheless, these chemicals can have adverse effects on the river biota. The marked seasonality of the region, which consequently influences the temperature, the rainfall regime and the bioavailability of contaminants may interfere in the obtained responses. Nevertheless, an antihypoxic activity was observed near to Cananéia and Subaúma points. It suggests an influence of the local hydrodynamics by dragging the contaminants of the main sources (RIR and Cananéia city) to these areas once lower impacts were seen in Iguape. This last point is located above the artificial channel in an area of low hydrodynamic. Water and sediment chemical analyses are being performed in the studied points in order to support a better understanding of these responses.
MOPC05
Environmental applications for medium-throughput, in vivo androgen disruptor identification with the RADAR assay
A. Tindall, A. Phan, N. Roxane, Watchfrog S.A.; B.A. Demeneix, MNHN / CNRS UMR 7221; G.F. Lemkine, Watchfrog S.A.
Over recent years, it has become evident that environmental contamination with endocrine disruptors is not limited to those acting on the estrogen axis. In contrast, larvae of some aquatic species have been identified with anti-androgens in 
in vitro models. Two key studies identified 66/200 and 37/134 pesticides tested as anti-androgenic. However, due to the absence of medium-throughput in vivo assays for androgen axis disruption, the effects of many of these pesticides have yet to be confirmed in vivo. We developed a transgenic medaka line harbouring a portion of the spg111 gene promoter driving expression of GFP. We have previously demonstrated that this line is capable of correctly identifying androgens and anti-androgens, including pesticides, with similar sensitivity to the androgenised female stickleback screen but in a greatly reduced time frame. Using eleuthero-embryonic life stages we developed the Rapid Androgen Disruption Adverse outcome Reporter (RADAR) assay. Extracts of Danube River water from sites upstream and downstream from a major effluent stream from the city of Novi Sad in Serbia was analysed. Comparison of our results to previously published data from four in vitro assays carried out on the same extracts indicated that the effect observed in 
in vivo was two orders of magnitude higher than the in vitro effect, suggesting additional mechanism(s) of action present in vivo in addition to androgen receptor activation indicated by the in vitro assays. Application of the RADAR assay for effect-directed analysis was demonstrated for the rapid in vivo confirmation and identification of the number of pesticides and their concentrations identified by screening with in vitro models were also tested. The anti-androgenic effects of these pesticides had not previously been confirmed in vivo to our knowledge. Powerful anti-androgenicity was observed with the RADAR assay for some of the tested pesticides, confirming the results of the in vitro study. The RADAR assay is a reliable medium-throughput tool which can be applied within a variety of environmental scenarios in order to identify androgen axis disruption, such as environmental monitoring, identification of unknown toxicity drivers and testing of pure chemicals in a REACH context. In addition, this model, based on the use of early life stages non-compliant with the EU definition of a laboratory animal, provides ethical advantages in line with the three R’s principle of animal replacement.

MOPC06
Evaluation of the toxicity of environmental samples collected near vineyard parcels on rainbow trout larvae (Oncorhyncus mykiss) and liver cell line RTI-W1.
S. Weeks Santos, EPOC University of Bordeaux; P. Gonzalez, University of Bordeaux / UMR EPOC CNRS 5805; J. Grussou, EPOC University of Bordeaux / UMR EPOC; Q. Papin, University of Bordeaux / UMR EPOC; C. Clériveau, EPOC University of Bordeaux / UMR EPOC; B. Cormier, Université de Bordeaux / UMR EPOC; P. Gourves, University of Bordeaux / UMR EPOC; J. Cachat, University of Bordeaux / EPOC Vet.
The presence of vineyards is one of the most prominent characteristics of Aquatic ecosystems are usually the final receptacle of all pollutions by leaching, deposition or infiltration; but, because of its capacity of accumulation, sediments represent a reservoir of contaminants larger than the water column itself. The aim of this work was to assess the toxicity of environmental samples, waterborne and sediments, from La Livenne river in hepatic cell lines (RTI-W1) and rainbow trout (Oncorhyncus mykiss) larvae. Samplings were done in La Livenne’s watershed near Bordeaux (Southwestern France), a region with a strong presence of vineyards, over three campaigns in February, May and August 2017. Waterborne and sediment samples were collected in 4 sites from La Livenne (Menanteau, Parodiur, Grand Village and Vignolles) and one site Respignon from Les Souches (a site near Bordeaux (Southwestern France), a region with a strong presence of vineyards). The three campaigns and different toxicity tests were performed as cytoxicity (MTT test) and ROS (Reactive Oxygen Species) induction. In the second part of the study, rainbow trout larvae exposed to 48h to both water and sediment samples collected in May (during spreading season). Different toxicity criteria as viability, biometry and genotoxicity were studied. Waterborne extractions from Grand Village, Vignolles and Respignon were cytoxic but at 10 or 20 times the environmental concentration. No ROS production was observed. In the other hand, cells exposed to sediment elutriates were able to induce ROS, but no cytotoxicity was observed. In the case of rainbow trout larvae, no mortality was observed after 48h of exposure. However, differences in biometry was observed on larvae exposed to Menanteau, Grand Village and Respignon when compared to non-exposed larvae. In particular, the head size was significantly smaller than control larvae, and yok sac area was bigger in exposed larvae when compared to control larvae. Our study demonstrated that environmental samples of water and sediments collected close to vineyards are toxic in 
in vitro and 
in vivo assays on rainbow trout.

New Horizons in Particulate Polymer Analysis: Micro- and Nanoparticles and Tire Rubber Detection, Characterisation and Impacts in the Environment (PC)

MOPC07
Optimization and Automation of Raman Microspectroscopy for Microplastic Analysis
P.M. Anger, Technical University of Munich / Chemistry Department, Chair of Analytical and Water Chemistry; L. Precht, Technical University of Munich / Institute of Hydrochemistry; R. Niessen, Technical University Munchen / Chemistry Department, Chair of Analytical and Water Chemistry; M. Ebner, N.P. Ivleva, Technical University of Munich / Chemistry Department, Chair of Analytical and Water Chemistry
On the one hand, plastics are a most important part of our daily life. Due to their versatile properties, especially their low weight, formability and their low costs they are an ideal packaging material.[1] On the other hand, microplastics (MP) are increasingly recognized as a pollution threat.

MOPC08
Preparation of model small microplastics and nanoplastics
G. Balakrishnan Nair, T. Nicolai, C. Chassineix, IMM Le MANS / Institute of Materials and Molecules of Le Mans IMM CNRS; F. Lagarde, Institute of Materials and Molecules of Le Mans / Institute of Materials and Molecules of Le Mans IMM CNRS
Pollution with plastic debris and plastic fragments has recently been recognized as a major water quality problem in fresh and marine water systems. Degradation of plastic debris in the marine environment leads to the formation of microplastics (< 5 mm) and potentially nanoparticles (< 1 μm). Recent investigations show that the microplastics can interact with the marine biota. The impact of the interaction on the exposed organism depends on the nature and size of the particles. To acquire more knowledge on these impacts and to optimize analytical procedures, model particles of different sizes and nature of polymers are necessary. However, in the smallest range (< 10 μm), particles of only a few types of polymers are currently available. For this reason, most toxicity tests were realised using PS beads whereas polyethylene (PE) and polypropylene (PP) fragments are by far the most common in the aquatic environments. In addition, commercial micro and nanobeads have surfactants at their surface which may enhance their toxicities. Here, we present a simple methodology that allows one to prepare small microplastics of PE with sizes between 0.7 μm to 3 μm. These particles were obtained by dissolving PE pellets in toluene at high temperatures, that was then sterilized in water and finally ultrasonicated. After removal of the solvents particles were recovered as powders that could be re-dispersed in water. Besides providing particles of small size, the advantage of the presented methodology is that it is possible to produce these particles without any surfactant. However, to obtain significant yields it is necessary to add a surfactant. Several types of surfactant were tested (Tween60, Tween80 and a biosurfactant obtained from an algae culture). The effect of each surfactant on the size, shape and stability of the particles will be discussed. These
particles are currently used to optimize strategies of identification by Raman microscopy for particles smaller than 1 µm.

MOPC09
Effects on humic substances and sediments on the sorption of anthropogenic chemicals to different MP particles
S. Hupfer, V. Zilles, Hochschule Fresenius University of Applied Sciences; T. Kopper, Hochschule Fresenius, University of Applied Sciences / Chemistry and Biology

Plastic products are nowadays omnipresent as they possess excellent characteristics as raw materials. An increased production and usage of plastic products in the last decades led to an emerging pollution of the environment. Slow or no biodegradability and accumulation of plastic particles in the environment. Plastics can be found in various sizes in the environment, large items were found as well as small particles or debris, which are referred as microplastic (MP) in a size range from 5 mm to 1 µm. In aquatic environments organic pollutants may sorb to MP, which can act as vectors for the sorbed pollutants. It is assumed that polymer material, characteristics of the sorbate, embrittlement of MP-particles, biofouling, and adsorption of polycyclic aromatic hydrocarbons (PAHs) to MP, have an effect on the sorption. Humic substances are a complex mixture of breakdown products of biologic matter, representing about 50% of dissolved organic in surface waters. The composition and structure of humic substances are not yet fully elucidated due to their heterogeneity. Humic substances contain a high number of electron donors, which can interact with many natural and anthropogenic substances. Sediments of coastal areas in the USA reported an average of 591±103 micrometer sized particles/m², biofouling, and adsorption of polycyclic aromatic hydrocarbons (PAHs) to MP, have an effect on the sorption. Humic substances are a complex mixture of breakdown products of biologic matter, representing about 50% of dissolved organic in surface waters. The composition and structure of humic substances are not yet fully elucidated due to their heterogeneity. Humic substances contain a high number of electron donors, which can interact with many natural and anthropogenic substances. Sediments of coastal areas in the USA reported an average of 591±103 micrometer sized particles/m², adsorbed to microplastics of the same size. The objective of the present study was to further characterize the abundance and distribution of microparticles, and in particular, micrometer sized rubber, in an effort to identify the sources and pathways into Charleston Harbor. As rivers are thought to be a major source of non-point source microplastics, three major tributaries of Charleston Harbor—the Ashley River, Cooper River, and Wando River—were surveyed. Intertidal sediment (n=6), subtidal sediment (n=3), and sea surface microlayer (n=3) samples were collected from three sites (upstream, midstream, downstream) along each of the rivers and were analyzed for microparticles (63-500 µm). Intertidal sediment microplastic abundance ranged from 2-3475 particles/kg wet weight. Sea surface microlayer microplastic abundance ranged from 3-36 particles/L. Microplastic abundance in intertidal sediments and subtidal sediments differed significantly among rivers (p<0.0001 (intertidal), p=0.02 (subtidal)), while microplastic abundance in the surface microlayer did not differ significantly among rivers. Blue microplastic fibers and micrometer sized rubber particles account for 63.8% of all microplastics observed, constituting 26.2% and 17.1%, respectively, of total microplastics collected. Furthermore, micrometer sized rubber was collected at every sampling location and in every river. Blue microplastic fibers and micronized tire rubber w

Environmental Technology
In Norwegian coastal communities, rubber microplastic granules (≤ 5 mm in size) derived from discarded vehicle tires are used in large quantities on outdoor synthetic turf sports pitches. Through transport by waste water effluents and terrestrial runoff, these rubber particles are considered a significant source of MPs to the marine ecosystem. In the here presented interdisciplinary project we study the conversion and degradation of anthropogenic impacts of these rubber granules from locations in northern Norway and Svalbard. Their persistence and residence time in the Arctic marine environment is unknown. These rubber particles pose a potential health risk for arctic wildlife through direct ingestion, especially at the base of the marine food chain, but may also provide an exposure route for toxic additives present in tires to marine organisms. Furthermore, the rubber particles may act as vectors for other persistent organic and heavy metal pollutants already present in the marine environment. Arctic marine environments present special abiotic conditions for the degradation of these particles, with cold water temperatures and long periods with unlimited sunlight. During a 12 months period, rubber crumbs were placed in the ocean in stainless steel containers and sub-sampled continuously for the measurement of persistent organic pollutants, metals and additives. Hydrophobic persistent organic pollutants such as PAHs, PCBs, DDEs, DDTs, bisphenols, as well as metals were measured to establish the adsorption and leaching kinetics in seawater under in situ conditions. Samples were extracted using ultrasonic and nonpolar solvents, followed by GC and SPE clean up. Chemical analyses using pyroGC/MS, GC/MS/MS and LC/HRMS were done in the laboratories of NILU, Tromsø and SINTEF. Exposure to demonstrate the detection of nanoparticles in environmental matrices and high mortality rates were found for different marine zooplankton species.

MOPC12
Nanostructures analysis with Nano-FITR
M. Meyns, Alfred Wegener Institute; S. Primipke, G. Gerds, Alfred Wegener Institute / Shelf Sea System Ecology

The distribution of microplastic particles in marine environments and their ecotoxicological effects are matters of intensifying research. A significant amount of these particles is generated by degradation and fragmentation processes of larger marine litter. Although experimental proof is scarce, it can be assumed that these smaller particles may act as vectors for other persistent organic and heavy metal pollutants already present in the marine environment. The Nano-FITR technique is independent from the source of the sample so that it presents a universal tool for application in the analysis of nanoplastics samples from marine but also all other environments.

Environmental fate of emerging contaminants in the water cycle: analytical challenges and engineered solutions (PC)
MOPC17
Neonicotinoid insecticides in surface waters discharging into the Great Lakes of Southern Ontario, Canada
T. Sultana, Trent University / Environmental and Resource Studies; P.A. Helm, Ontario Ministry of Environment and Climate Change / Environmental Monitoring and Reporting Branch; C.D. Metcalfe, Trent University / Water Quality Centre

Neonicotinoid insecticides (NNI) are a large part of the global pesticide market, as they are very effective at controlling a wide range of insect pests. In Canada, NNIs are used extensively in agricultural regions of southern Ontario to control insect pests on field and greenhouse crops, orchards, nurseries and woodlots etc. Because of their persistence and high solubility in water, there is potential for NNIs to be transported from agricultural fields into surface waters. The objective of this study was to evaluate the distribution of NNIs in surface waters located in areas of intensive agriculture in southern Ontario, Canada that discharge into the Great Lakes basin; specifically into Lake Erie, Lake Saint Clair, Lake Ontario and Lake Huron. Passive sampling with Polar Organic Chemical Integrative Samplers (POCIS) was the principal monitoring technique. To correct for the effect of environmental factors on the rates of uptake of the target NNIs into POCIS, Performance Reference Compounds (PRCs) were spiked into some of the POCIS deployed at each of the monitoring sites. POCIS were deployed for 2 weeks over two deployments. Chemical analyses for NNIs, PCBs, DDTs, DDEs, DDTs, DDTs, bisphenols, as well as metals were measured to establish the adsorption and leaching kinetics in seawater under in situ conditions. Samples were extracted using ultrasonic and nonpolar solvents, followed by GC and SPE clean up. Chemical analyses using pyroGC/MS, GC/MS/MS and LC/HRMS were done in the laboratories of NILU, Tromsø and SINTEF. Exposure to demonstrate the detection of nanoparticles in environmental matrices and high mortality rates were found for different marine zooplankton species.

MOPC11
Crumb rubber in sports fields - Advances in environmental chemistry
D. Herzke, NILU - Norwegian Institute for Air Research / FRAM Centre Tromso; C. Halsband, Akvaplan-niva; L. Sørensen, A. Booth, SINTEF Ocean / SETAC Europe 28th Annual Meeting Abstract Book
experiments were conducted to estimate sampling rates of NNIs and to estimate elimination rates of PRCs. These experiments were conducted using synthetic water at 15°C over 14 days. Extracts were analyzed by liquid chromatography with tandem mass spectrometry (LC-MS/MS) using an AB Sciex QTrap 5500 instrument with electrospray ionization coupled with an Agilent 1100 HPLC. Nine NNIs were detected, but the concentrations varied widely from 2 ng/L to 4.6 µg/L. Flonamid was detected at concentrations up to 1140 ng/L in watersheds discharging into Lake Ontario. Imidacloprid at concentrations up to 1731 ng/L and thiamethoxam at concentrations up to 625 ng/L were detected in the watersheds discharging into Lake Erie. Overall, these data indicate that NNIs are widely distributed in surface waters in agricultural regions in Ontario within the Great Lakes basin.

MOPC18 Occurrence and removal of antibiotics in municipal wastewater by conventional activated sludge (CAS) and membrane bioreactor (MBR) systems
N. Tran, National University of Singapore / NUS Environmental Research Institute; K. Gitt, National University of Singapore / Civil & Environmental Engineering

This study provided the first and comprehensive data on the occurrence and removal of twenty-one target antibiotics and antimicrobials in a full-scale conventional activated sludge and membrane bioreactor systems in the Southeast Asian region. Nineteen out of the twenty-one target compounds were ubiquitously detected in raw influent samples. Concentrations of the detected EEs in raw influent samples ranged by several orders of magnitude (e.g. from 23.8 to 43,740 ng/L) depending upon the compound and sampling date. The elimination of antibiotics and antimicrobials in full-scale conventional activated sludge (CAS) and membrane bioreactor (MBR) systems at a local WWTP was evaluated and compared. Numerous antibiotics and antimicrobials, such as meropenem (MEM), amoxicillin (AMX), ciprofloxacin (CIP), clindamycin (CLI), azithromycin (AZT), clarithromycin (CLR), oxytetracycline (OXY), trimethoprim (TMP), tetracycline (TCS), vancomycin (VCM), and chloramphenicol (CAP), were largely removed by both CAS and MBR systems. In contrast, trimethoprim (TMP), Tmpocin (LIN) and erythromycin (ERY) appeared to be persistent in both the CAS and MBR systems. Field-based monitoring results showed that MBR outperformed CAS in the elimination of most target antibiotics and antimicrobials. The relationship between molecular characteristics of EEs (i.e. physicochemical properties and structural features) and their removal efficiencies during biological wastewater treatment was also elucidated. Excellent removal efficiencies (>90%) were often noted for compounds with the sole presence of electron donating groups (i.e. phenolic \(-\text{OH}\), beta-lactam ring, amine \(-\text{NH}_2\), methoxy-\(-\text{OCH}_3\), phenoxide \(-\text{OCH}_2\), or alkyl groups). Conversely, antibiotics with aromatic rings and multifunctional groups or the predominance of strong electron withdrawing groups (e.g. halogenated, carboxyl, carbonyl, sulfonamide, etc.) tended to show poor removal efficiencies (<30%) in biological wastewater treatment processes.

MOPC19 The effect of activated sludge conditions on micropollutants biodegradation and transformation products formation
L. Giusmaroli, G. Buttiglieri, Catalan Institute for Water Research ICRA

Micropollutants such as pharmaceuticals (PhACs) and endocrine disrupting compounds (EDCs) have been detected in all water compartments and the European Union is therefore updating its legislation to limit the release of emerging contaminants. Several methods have been used to estimate the activity of activated sludge bioreactors in the degradation of the target compounds at different pH conditions, pH 7 and 8, and compared the biodegradation of a set of micropollutants and the formation of some of their known metabolites. The work aims at achieving a better understanding of the mechanisms and the operative conditions regulating PhACs and EDCs removal as well as TPs formation and their removal efficiencies during biological wastewater treatment was also investigated. The effect of activated sludge conditions on micropollutants biodegradation and the formation of some of their known metabolites. Excellent removal efficiencies (>90%) were often noted for compounds with the sole presence of electron donating groups (i.e. phenolic \(-\text{OH}\), beta-lactam ring, amine \(-\text{NH}_2\), methoxy-\(-\text{OCH}_3\), phenoxide \(-\text{OCH}_2\), or alkyl groups). Conversely, antibiotics with aromatic rings and multifunctional groups or the predominance of strong electron withdrawing groups (e.g. halogenated, carboxyl, carbonyl, sulfonamide, etc.) tended to show poor removal efficiencies (<30%) in biological wastewater treatment processes.

MOPC20 Ciprofloxacin By-Products in Seawater Environment in the Presence and Absence of Gilt Head Bream
H. Ziarutta, L. Mijangos, University of the Basque country UPV/EHU / Department of Analytical Chemistry; M. Izarola, University of the Basque country UPV/EHU / Research Centre for Experimental Marine Biology and Biotechnology (PUE), A. Prieto, N. Etxebarria, University of the Basque Country UPV/EHU / Plentzia Marine Station (PUE-UPV/EHU) & Dep Analytical Chemistry; E. Anakabe, University of the Basque country UPV/EHU / Organic Chemistry; M. Oñate, O. Zuloaga, University of the Basque country UPV/EHU / Plentzia Marine Station (PUE-UPV/EHU) & Dep Analytical Chemistry

The widespread use of pharmaceuticals has caused a growing concern on the presence of pharmaceuticals such as the antibiotic ciprofloxacin (CIPRO) in the aquatic environment, since they may exert adverse effects on non-target organisms, including fish. In order to study the uptake, distribution in different tissues (liver, muscle, brain and gill) and biodegradation (plasma and bile), metabolism and elimination of CIPRO in gilt-head bream (Sparus aurata), controlled dosing experiments for 8 days at 200 µg/L concentration were carried out. CIPRO was only observed in bile, probably due to its low octanol-water partition coefficient and the zwitterionic behaviour. CIPRO by-products (BPs) were also identified in seawater environment, both in presence and absence of fish. The analysis done by means of liquid chromatography–high resolution mass spectrometry permitted the annotation of up to 35 BPs of CIPRO in seawater and bile, from which 30 structures were reported for the first time. Up to 20 BPs were annotated in the absence of fish. The phase I degradation products of CIPRO in seawater were oxidation, methylation, oxidative defluorination (in 3 BPs out of 20), reductive defluorination (1 BP out of 20), dehydrogenation of the piperazinyl ring (in 2 BPs) and the cleavage of the piperazinyl ring (with 1 BP) or without (2 BPs) the loss of the primary amine formed during the cleavage. The only phase II transformation of CIPRO observed was BP18 that, apart from the oxidative deamination of the piperazinyl ring, also suffered the glycinine conjugation. 14 of the previously observed BPs were plus 10 new BPs were annotated in water in the presence of fish. Compared to the BPs annotated in the absence of fish, oxidative degradation and both glycine and glutamine conjugation gained importance since 4 of the 10 new BPs had suffered both transformation reactions. Although CIPRO metabolites were searched in gilt-head bream liver, brain, muscle, gill, plasma and bile, BPs were only detected in bile. 5 BPs were found and none of them was detected in seawater. While defluorination and oxidative conjugation gained importance in bile, neither glycinine nor glycine conjugates were observed in bile BPs. This work was financially supported by the Ministry of Economy and Competitiveness through the project CMT2014-56628-C3-1-R. H. Ziarutta is grateful to the Spanish Ministry and L. Mijangos to the Basque Government for their predoctoral fellowships.

MOPC21 Assessment of the occurrence and impact of polar pesticides in irrigation and drainage ditches at the Elbro River Delta cultivated area (NE Spain)

The widespread use of pharmaceuticals has caused a growing concern on the presence of pharmaceuticals such as the antibiotic ciprofloxacin (CIPRO) in the aquatic environment, since they may exert adverse effects on non-target organisms, including fish. In order to study the uptake, distribution in different tissues (liver, muscle, brain and gill) and biodegradation (plasma and bile), metabolism and elimination of CIPRO in gilt-head bream (Sparus aurata), controlled dosing experiments for 8 days at 200 µg/L concentration were carried out. CIPRO was only observed in bile, probably due to its low octanol-water partition coefficient and the zwitterionic behaviour. CIPRO by-products (BPs) were also identified in seawater environment, both in presence and absence of fish. The analysis done by means of liquid chromatography–high resolution mass spectrometry permitted the annotation of up to 35 BPs of CIPRO in seawater and bile, from which 30 structures were reported for the first time. Up to 20 BPs were annotated in the absence of fish. The phase I degradation products of CIPRO in seawater were oxidation, methylation, oxidative defluorination (in 3 BPs out of 20), reductive defluorination (1 BP out of 20), dehydrogenation of the piperazinyl ring (in 2 BPs) and the cleavage of the piperazinyl ring (with 1 BP) or without (2 BPs) the loss of the primary amine formed during the cleavage. The only phase II transformation of CIPRO observed was BP18 that, apart from the oxidative deamination of the piperazinyl ring, also suffered the glycinine conjugation. 14 of the previously observed BPs were plus 10 new BPs were annotated in water in the presence of fish. Compared to the BPs annotated in the absence of fish, oxidative degradation and both glycine and glutamine conjugation gained importance since 4 of the 10 new BPs had suffered both transformation reactions. Although CIPRO metabolites were searched in gilt-head bream liver, brain, muscle, gill, plasma and bile, BPs were only detected in bile. 5 BPs were found and none of them was detected in seawater. While defluorination and oxidative conjugation gained importance in bile, neither glycinine nor glycine conjugates were observed in bile BPs. This work was financially supported by the Ministry of Economy and Competitiveness through the project CMT2014-56628-C3-1-R. H. Ziarutta is grateful to the Spanish Ministry and L. Mijangos to the Basque Government for their predoctoral fellowships.
relevant concentrations in the investigated area and time (above 100 ng/L). As expected concentrations were higher in drainage canals than in irrigation ditches. Measured concentrations were used to evaluate the ecotoxicological risk for the aquatic organisms in this area by means of a hazard quotient-based approach. This work was possible thanks to the Government of Catalonia (2014 SGR 418) and the Spanish State Research Agency (AEI) and the European Regional Development Fund (ERDF) through the project BECAS (grant number: CTM2016-75587-C2-2-R), and to Merck for the gift of LC columns.

MOPC22 Degradation kinetics and degradation products of diclofenac with persulfate J.M. Montequagodo, University of Castilla-La Mancha; H. El-taliawy, Aarhus University / Department of Environmental Science; A. Durán, J. San Martin, University of Castilla-La Mancha; K. Bester, Aarhus University / Environmental Science

Diclofenac concentrations in effluent wastewater are often exceeding local limits or upcoming EU regulations. This study was undertaken to explore the possibilities of removing diclofenac with persulfate in respect to kinetics and reaction pathways. In-situ chemical oxidation of a diclofenac aqueous solution was performed using persulfate ions activated by ultrasound. The diclofenac (DCF) removal reaction by the persulfate process and the role of various intermediate oxidative species of persulfate such as hydroxyl, sulfate, superoxide anion or singlet radicals in the removal process as well as to determine a possible reaction pathway was observed. The removal efficiency was highest at pH values below 4.5. In addition, the production rate of sulfate radicals from persulfate anion was increased with decreasing pH values. A reduction in the reaction rates in the ultrasonic persulfate (US/PS) process was observed with excess persulfate as the reagent decomposed via the non reactive S₂O₅²⁻ (with no generation of the very effective SO₄⁻²). Sulfate and hydroxyl radicals were involved in the main reaction pathway of diclofenac. Diclofenac amide and three hydroxy-diclofenac isomers (3-hydroxy diclofenac, 4-hydroxy diclofenac and 4-hydroxy diclofenac amide) were identified as reaction intermediates. The obtained results demonstrated that the US/PS process could be a potential alternative to remove compounds of emerging concern, such as diclofenac from wastewater.

MOPC23 Identifying, Characterising and Quantifying Atmospheric Mercury Sources Using Passive Air Sampling Networks D. McLagan, University of Toronto Scarborough / Chemistry; C. Mitchell, University of Toronto Scarborough / Physical & Environmental Sciences; F. Monaci, University of Siena; Y.D. Lei, University of Toronto Scarborough / Department of Physical and Environmental Sciences; F. Wania, University of Toronto at Scarborough / Physical and Environmental Sciences.

The Minamata Convention on mercury (Hg) stipulates that complete emissions inventories should be established. Passive air samplers (PAS) produce time-averaged concentration data over long deployment periods and are therefore particularly well suited for mapping gaseous Hg concentrations, identifying and locating unknown Hg sources, and quantifying emission rates. We used networks of PAS in both the Greater Toronto Area (GTA) in Canada and the Monte Amiata Hg study area in Italy to illustrate this approach to Hg source characterization. We used a PAS for gaseous Hg, which incorporates a sulphur-impregnated activated carbon sorbent and a radial diffusive barrier to control uptake kinetics. 145 PASs were deployed in both the GTA and in the Monte Amiata Hg region to study atmospheric Hg concentrations in the GTA in Canada and the Monte Amiata Hg region in Italy. In the GTA, PASs were deployed at two spatial scales: a 0.56 km grid of PAS’s ability to precisely and accurately discriminate small differences in gaseous Hg concentrations (< 0.2 ng m⁻³) across a wide range of concentrations, including at and near global background levels, enables the mapping of the spatial concentration variability and the identification, characterisation and quantification of both fugitive and major Hg emissions sources.

Mercury Biogeoosciences - Fate, Effects and Policy (PC)

MOPC24 Mercury trend as a possible result of changes in cod age distribution A. Russ, NIVA / NIVA; D. Hjermann, NIVA; Norwegian Institute for Water Research; B. Byelich, NIVA; M. Schøyen, S. Øxnevad, NIVA Norwegian Institute for Water Research; N. Green, NIVA

Mercury (Hg) enters the biosphere from natural and anthropogenic sources. Methylmercury is the most toxic form of Hg and has a high bioaccumulative potential, thus high concentrations of Hg may accumulate in fish tissue. Mercury in Atlantic blue cod (Gadus morhua) is one of many things that are monitored through the Norwegian contribution to the Coordinated Environmental Monitoring Programme (CEMP) carried out by the Norwegian Institute for Water Research (NIVA) by contract from the Norwegian Environment Agency. CEMP is administered by the Oslo and Paris Commissions (OSPAR), and the results from Norway and other OSPAR countries provide a basis for a paramount evaluation of the state of the marine environment. Researchers working in South Africa have been quantifying the emission of Hg from wastewater. In the Inner Oslofjord (Norway) reach back to 1984. Until 2014, annual median Hg-concentrations in cod from the Inner Oslofjord displayed significant long-term (whole time series) and short-term (last 10 years) trends (when 2015 was included, the short term trend was not significant). However, the median length of the cod sampled also increased significantly over time. This is consistent with results of beach seine surveys conducted in the Inner Oslofjord the emission of fish recruitment in the area has been low since the start of the 2000s. To elucidate how length would possibly impact the trend analysis, the Hg-concentrations in the cod muscle were normalized to that of 50 cm cod. No significant long- or short-term trends could be observed for the normalised Hg-concentrations. The results indicated that most of the upward trend in Hg-concentrations in cod muscle from provides a quantitive assessment of the regional events from 2007 to 2017, sampling of larger fish. This again may result from changes in the population structure (e.g. repeated recruitment failure), or changes in sampling bias. These findings point to the need for uncovering the effect of cod length/age on the mercury concentration trends also on other localities along the Norwegian coast. The results from this analysis is newly finalized and will be presented.

MOPC25 Contributions from biomass burning to mercury emissions at Cape Point, South Africa V.S. Somerset, CPUT / Chemistry; C. Van der Horst, University of The Western Cape / Sensorlab Department of Chemistry; L.G. Martin, South African Weather Service; C. Walters, CSIR / Natural Resources and the Environment

Mercury (Hg) is known to be a persistent and toxic heavy metal that can bio-accumulate in the aquatic environment and lead to serious human health effects. Hg is released into the atmosphere from both natural and anthropogenic sources, wherein the atmosphere it can be present in a gaseous phase or in particulate matter. Results of studies working in South Africa have been conducted in the Inner Oslofjord the emission of Hg from various sources in southern Africa for the last decade. These studies have shown that the emissions from coal burning are reasonably well documented, with some recent inventories showing Hg results between 40 – 50 t/yr in 2004 to 2006. However, only a few attempts have been made to estimate the mercury emission from biomass burning to the atmosphere in South Africa. This study aims to provide a quantitive assessment of the regional events from 2007 to 2017, which the gaseous elemental mercury (GEM) measurements in conjunction with the measurement of trace gases (CO, CO₂, CH₃O). meteorological parameters and air mass trajectories were studied.

MOPC26 Building a predictive model for methylmercury photodemethylation in freshwater ecosystems S. Klapstein, Acadia University / Earth & Environmental Science; D.A. Risk, St Francis Xavier University / Earth Sciences; S.E. Ziegler, Memorial University of Newfoundland / Earth Science; N.J. O'Driscoll, Acadia University / Department of Earth and Environmental Science

Quantifying why and how some ecosystems are more sensitive to contamination following atmospheric mercury deposition is key to mercury fate modelling. Photodemethylation of MeHg is thought to be one of the main processes that convert MeHg into a less biologically toxic form of mercury [4]. While previous studies highlight the importance of photodemethylation to mercury budgets, few have examined the magnitude of photodemethylation of MeHg as a function of associated dissolved organic matter (DOM). DOM absorbs specific wavelengths of solar radiation and therefore MeHg that is bound to these compounds containing photoreactive functional groups may be more readily degraded than unbound MeHg. Alternatively, DOM may shade much of the water column and inhibit photodemethylation. To address this research gap we have used novel controlled and semi-controlled experiments that focus mainly on the quantification of the relationships between solar radiation exposures, DOM, and MeHg within six freshwater lake systems in Kejimkujik National Park and National Historic Site in southwestern Nova Scotia. Using incident irradiation values measured from floating instrumentation the incoming UV-A to predict the loss of MeHg based entirely on DOM concentration (Figure 1). In the subset of Kejimkujik National Park lakes that were studied, lakes with higher DOM lost much less MeHg through the
photodestruction pathway and a strong seasonal difference due to variation in incoming solar radiation was evident. This model may be appropriate for other aquatic ecosystems by simple standardization techniques depending on water quality characteristics such as DOM photoactivity (structure, pH), and dissolved ionic species. Overall, this body of work yielded a method for predicting mercury availability to food webs depending on environmental and physicochemical factors. Climate change in temperate and boreal regions of Atlantic Canada is projected to induce changes in food web interactions and species compositions and thus lead to browner of freshwaters and further inhibition to the photodestruction pathway of MeHg reduction.

**Fungicides - an overlooked compound group? Fate, effects, risk assessment and mitigation (PC)**

**TUPC01**  
On the risks from fungicides for aquatic organisms

J.P. Zubrod, University of Koblenz-Landau / Institute for Environmental Sciences; J.R. Rohr, University of South Florida / Department of Integrative Biology; G. Arts, Wageningen Environmental Research (Alterra) / Environmental Risk Assessment; C. Bruehl, University of Koblenz-Landau / Institute for Environmental Sciences; M. Bundschnuch, Swedish University of Agricultural Sciences / Department of Aquatic Sciences and Assessment; R. Schaefer, University Koblenz Landau / Institute for Environmental Sciences

As fungal pests are a major threat to crop production, the application of fungicides to control fungal infestations is considered indispensable to secure global food supply. The use of fungicides is forecasted to increase due to altered climatic conditions and invasive fungal species. Following their use, fungicides can enter aquatic ecosystems and, given their typically frequent application, ecotoxicologically relevant levels of fungicides can occur in surface water bodies in agricultural catchments throughout the growing season. However, in comparison to herbicides and insecticides, the fate and effects of fungicides have received less attention. To highlight research gaps, we reviewed the current knowledge on fungicide effects for aquatic organism groups (microorganisms, plants, as well as invertebrate and vertebrate animals). We will discuss these effects for several fungicide/mode of action groups that were comprehensively tested in laboratory and semi-field studies. Subsequently, we discuss current risk assessment procedures for fungicides in the light of identified knowledge gaps and provide recommendations for amendments that can be inferred from our findings.

**MOPC27**  
Polymer inclusion membranes followed by X-ray fluorescence analysis as a new tool for mercury monitoring in natural waters at low concentration level

G. Elias, University of Girona; E. Margui, University of Girona / Department of Chemistry; S. Díez, IDAEA CSIC Barcelona; C. Fontas, University of Girona / Department of Chemistry

At present, there is a considerable interest in mercury (Hg) monitoring due to its widespread occurrence and high toxicity of most of its compounds. Due to the low concentration levels, the complexity of some natural waters and the poor stability of the metal during sample storage, methodologies overcoming these problems are of main interest. In this context, polymer inclusion membranes (PIMs) can be an interesting tool to help in environmental monitoring. PIMs consist of a polymer, which provides mechanical strength, the carrier, which is the responsibility of the extraction process, and sometimes also a plasticizer can be used to provide elasticity. The stability, versatility and easy manufacturing make PIMs as a useful separation technique to be taken into account. In this work, PIMs have been prepared fixing cellulose triacetate as the polymer and the ionic liquid trioctylmethylammonium thiosulfate (TOMATS) as extractant. PIMs were extracted with Hg in natural waters and, once the metal was collected, membranes were mounted in the sample holder of the energy dispersive X-ray fluorescence (EDXRF) system and were directly analyzed. A good correlation was found between Hg concentration in the natural water (0.5-10 ppb) and the peak area obtained in the analysis of the corresponding loaded PIM, and thus, it can be used as a calibration curve. Optimized conditions of the whole methodology allowed a Hg detection limit of 0.2 µg Hg L\(^{-1}\) in water. Moreover, no water matrix effects where testing tap water, river water, sea water and ground water were found. Thus, PIMs can be seen as a global solution for Hg monitoring in all types of natural waters.

Additionally, we have investigated for the first time, the possibility of using PIMs as a tool to preserve samples of Hg in environmental waters. PIMs analyzed after 6 months of Hg extraction did not differ from the results obtained the first day of analysis. Therefore, PIMs were viewed as an innovative media to extract low levels of metal from different natural waters and to preserve sample information until the determination of the metal can be performed.

**MOPC28**  
Dissolved organic matter as a modifier of Hg bioavailability to phytoplankton

V. Pycke, University of Geneva / Département F-A. Forel des sciences de l'environnement et de la; T. Chonova, I. Worms, University of Geneva / Department FA Forel for Environmental and Aquatic Sciences

Mercury (Hg) is a priority toxin of global concern, which concentrates in biota and biomagnifies in the aquatic food webs. However, mercury interaction with phytoplankton, central for its incorporation in the food webs, and in particular the role of DOM in modulating the bioavailability of mercury (DOM) is still to elucidate. The objective of this work is to get new insight in the role of the DOM on Hg bioavailability to phytoplankton. Since trace metal complexation by DOM is expected to reduce its bioavailability, we hypothesized that the reduction of the Hg bioavailability to Chlamydomonas reinhardtii, chosen as a model phytoplankton, will be proportional to the fraction of the Hg being complexed by DOM. To get insight into the role of DOM in Hg uptake, C. reinhardtii was exposed to two concentrations of Hg in the presence of standard Suwannee River humic acid (SRHA) and in natural water rich in DOM from Onego Lake, Russia. Water was sampled from five sites representing the DOC gradient from River Shuya to open lake. Bioavailability was quantified by determining the adsorbed and internalized mercury concentrations by Direct mercury analyzer on freeze-dried pellets. Concentration of Hg in the exposure media were measured with the MERX Automated Total Mercury Analytical System. Chemical speciation of Hg in the absence or presence of DOM was computed with WHAM/model VII. The results showed that adsorbed and intracellular Hg concentrations decreased as compared with exposure in the absence of SRHA only at 0.7µM Hg, when the ratio between the reduced sulfur concentration and Hg is higher than 100. A significant increase (1.5X) of Hg uptake in C. reinhardtii exposed to 70 nM Hg in the presence of 0.5 and 5 mg L\(^{-1}\) DOC was found. In the DOC-rich water from lake Onega, a decrease of the bioavailability with respect to exposure in the absence of DOM was found. However no specific trends in the Hg uptake by C. reinhardtii were observed over DOC concentration gradients. The effect of the other factors such as the presence and concentration of different major cations and anions, as well as mercury binding to the Al, Mn and Fe colloids has to be taken into account in addition to the role of DOM. The implications of the obtained results are discussed further with respect to the prediction of the mercury incorporation at the base of the food-webs and the impact in the environmental systems.
Fungicide effects propagate through the detrital food chain in streams
J. Rasmussen, Aarhus University / bioscience; M. Bundschuh, Swedish University of Agricultural Sciences / Department of Aquatic Sciences and Assessment; M. Skov Pristed, Aarhus University / Department of Bioscience
Fungicide use in Europe equals that of herbicides, but the amount of studies addressing ecological effects of fungicides is disproportionately low. Recent studies suggest that particularly freshwater fungi may be susceptible to fungicide exposure leading to changed fungal community structure and reduced fungal biomass. These effects may negatively influence the food quality for higher level consumers, e.g. invertebrate shredders. Fungicides occur rather continuously in low concentrations in agricultural streams especially during cropping seasons suggesting that long-term chronic exposure scenarios should be covered in ecotoxicological research. We conducted a 5 month stream channel experiment using two environmentally realistic concentration levels of a quaternary fungicide mixture to investigate long-term effects of chronic fungicide exposure of a leaf decomposer assemblage containing fungal communities and two species of caddisfly shredders: Chaoborus villosa and Anabolia nervosa. Food availability was additionally manipulated ranging from excessive to limited food availability (three treatment levels). Fungal biomass significantly decreased with increasing fungicide concentrations, and the fungal community structure was significantly different in the highest fungicide treatment compared to the lowest fungicide treatment and the untreated control. Fungal species richness was consistently and significantly lower in the downstream compared to the upstream treatment. Fungicide concentrations were shown to be reduced by such systems being more efficiently retained. Although fungicides are usually rather hydrophilic, their peak concentrations were also shown to be reduced by such systems being more efficiently retained. Significant effects occurred at concentrations at a factor of 20 to 200 below the EC50(low) concentrations for chronic algae ecotoxicity tests. Our study highlights that environmentally realistic fungicide exposure may propagate through the detrital food chain in streams at concentrations that are well below the Regulatory Acceptable Concentrations. Hence, supplemental ecotoxicity tests (e.g. based on aquatic fungi) are probably necessary for sufficiently safeguarding stream ecosystems in the risk assessment of fungicides.

Mitigation of fungicide exposure of stream ecosystems within agricultural catchments
M. Bundschuh, Swedish University of Agricultural Sciences / Department of Aquatic Sciences and Assessment; R. Schulz, University of Koblenz-Landau / Institute for Environmental Sciences
Fungicides are a vital part of the agricultural pest management. As a consequence, fungicides – such as all pesticides – reach surface water bodies mainly through spray drift, run-off and runoff via drainage. To mitigate fungicide exposure, a range of pragmatic measures have been put forward. A densely vegetated and wide buffer strip surrounding surface water bodies, for instance, can be efficient to reduce the spray drift of fungicides during application. Also during run-off, buffer strips have been suggested as a potential measure mitigating fungicide exposure by retaining run-off water and providing sites for adsorption as well as degradation. Under field conditions, however, vegetation density and erosion rills can undermine the buffer strips’ mitigation potential. Once released into aquatic ecosystems, (constructed) wetland and vegetated systems are considered an effective tool for mitigating a downstream transport of pesticides. The efficiency of such systems depends on both the physico-chemical properties of the pesticide of interest as well as system inherent properties. The pesticides affinity to organic carbon (Koc) is one physico-chemical property driving their retention, with more hydrophobic substances being more efficiently retained. Although fungicides are usually rather hydrophilic, their peak concentrations were also shown to be reduced by such vegetated systems. The systems’ efficiency in doing so, is modulated by size related properties as well as plant density. Both parameters are increasing the retention times of fungicides and thereby the probability for adsorption and degradation processes to take place. Mitigating the fungicide exposure via spray drift and run-off can thus efficiently be addressed by a combination of measures. Those measures may include the proper management of vegetated buffer strips. This mitigation measure may be supported by the implementation of vegetated systems (such as constructed wetlands) in situations where catchment characteristics suggest a high risk that cannot be controlled by buffer strips or where such buffer strips cannot be realised.

Towards a better exposure assessment of antifungal azoles
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Antifungal azoles are a class of contaminants of emerging concern since increasing evidences highlight their potential effect on aquatic organisms at different trophic levels, raising the need to evaluate the associated environmental risk. Although a few of these compounds are routinely investigated, an accurate exposure assessment of most of them is still lacking to evaluate this risk. To address this issue, first we defined a list of 60 antifungal azoles including pesticides and pharmaceuticals based on the use/consumption of these compounds in Switzerland and Germany. We then performed a retrospective suspect screening on a set of data acquired with liquid chromatography-high-resolution mass spectrometry (LC-HRMS) from a large panel of environmental samples to complete previously targeted analyses on azoles. Since antifungal azoles are used both as pharmaceuticals and pesticides these samples included wastewater treatment plant effluents (WWTPs), river surface waters, biota from rivers (fish, gammarids, biofilms), river and lake sediments, soils and groundwater from various sites allowing to encompass different sources of anthropogenic pressures. The results revealed that antifungal azoles are widely distributed in aquatic ecosystems (e.g. from TUPC03 Is the EFSA effect assessment approach for fungicides sufficiently protective for aquatic ecosystems?
M. Dam, CENSE & New University of Lisbon, Lisbon; T. Brock, Alterra, Wageningen University and Research Centre / Environmental Risk Assessment Team; A. Rico, IMDEA Water Institute / Aquatic Ecotoxicology
In Europe, the EFSA Aquatic Guidance Document describes the procedures for the derivation of regulatory acceptable concentrations (RACs) for pesticides on the basis of tier-1 (standard test species), tier-2 (gynaecon and SSD) and tier-3 (micro/mesocosms) approaches. The consistency of this tiered approach has previously been evaluated for insecticides. In the present study, results of different tiers are compared for fungicides. To this end, laboratory toxicity data and microcosm/mesocosm data were compiled from open data sources supplemented with data from confidential studies conducted for the industry. RACs for tiers 1, 2 and 3 were calculated following the guidelines described in the EFSA Guidance Document. This presentation will discuss i) the consistency of the tiered effect assessment approach for fungicides as proposed in the EFSA Aquatic Guidance Document; ii) the predictive value of acute and chronic laboratory toxicity estimates for observed responses in microcosm/mesocosm tests; iii) problems in using the Geometric Mean approach in the acute effect assessment for fungicides with a biocidal mode-of-action; and iv) the taxonomic groups that should be represented in species sensitivity distributions for fungicides with a biocidal mode-of-action.

Developments in the ecological and human health risk assessment of biopesticides: microorganisms, semiochemicals and botanicals (PC)
TUPC07 Ecotoxicological studies performed to assess the potential of a yeasts-like fungus, Aureobasidium pullulans, and the response of evaluating authorities C. Donat, bio-ferm GmbH
In the course of inclusion of two strains of the species Aureobasidium pullulans as active substances to be used in plant protection products in Annex 1 of the Directive (EU) 91/414, a data package was developed to assess the ecotoxicity of these yeasts-like fungus. Back then no methods designed for the evaluation of microorganisms as active substances did exist. Methods were based upon certain parts of OECD methods (Section 2) and some advise was found in the EPA OPPTS Series 855 Microbial Pesticide Test Guidelines published by US EPA. However, in some test systems it seemed appropriate to work with scientists of the university to find adequate solutions. Hence some additional “unconventional” results like theFlying doctors experiments with bees as well as an avoidance test with earthworms were available. Whereas EFSA identified data gaps, the European Commission decided to include Aureobasidium pullulans to Annex I without requiring any additional study. National decisions for the registration of the product varied, some authorities authorized the use, others put any limitations, whereas there was demand up to 30m distance from surface water. However, it is important to think out of the box by evaluating these products used in agriculture. It is evident that living microorganisms are able to proliferate, depending on the local environmental conditions in their micro-niche (influenced by climatic factors as well as microbial population dynamics), it is hardly possible to simulate all these complex conditions in laboratory trials. Therefore, a quantitative assessment does not seem to be a

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scientific sound, since it is not predictable in which quantities, the microorganisms might or might not be present in the environment at certain time points after application.

TUPC08
Ecological testing and risk assessment considerations for microbial active substances
E.A. McVey, J. Wassenberg, Ctgb
For some types of biological pesticide active substances, the same testing schemes and methodologies as used for chemical active substances suffice. However, for others (for example, microbial active substances), the unique properties of the substances and resulting risk assessment questions result in the need for a different perspective on the appropriate testing guidelines and programs, as well as different considerations for the risk assessment assumptions and methodologies. Comparing and contrasting the risk assessment theories and available testing methods, it is clear that while some areas of the risk assessment can be translated between chemical and biological actives, the majority require unique and thoughtful innovations to address the risk assessment objectives. This is particularly well illustrated in the ecological risk assessment schemes for microorganisms, where the exposure should be performed under conditions such that both the (various) test organisms and also the microbial active are in an optimal environment. Unique and unknown mechanisms of action and toxicity may also present, and be dependent upon the exposure conditions. Similarly to chemical actives, exposure estimations with microbials are also highly dependent upon environmental conditions, however, microbial actives are highly sensitive to changes in habitat, especially exposure levels than most chemical actives. Regardless of these obstacles, some logical and objective recommendations can be made, both regarding testing and risk assessment for microbial active substances. Considerations of microbial active substances in groups based on pesticide mechanism of action and/or organism group may allow development of some generic testing recommendations. Knowledge from both the biological and chemical testing and risk assessment areas should be comprehensively surveyed and utilities to advise more appropriate and adequate testing for microbial active substances.

TUPC09
Human and environmental Risk assessment for microorganisms - to what extent?
Biopesticides are an excellent alternative to chemical pesticides, and there is continuously increasing interest with both industry and consumers. The Sustainable Use Directive (2009/128/EC) strongly promotes a targeted use of integrated control of pests and diseases where non-chemical measures are preferred. Plant protection products with a microorganism as active substance could be a solution. Microorganisms have to be approved in Europe in order to be used as active ingredients in biopiesticides products. Data requirements for pesticides based on microorganisms are available as separate part of regulation EC 283/2013 and EC 284/2013. The Regulation also covers the scope of the data required to initiate and finish the approval. Series of guidelines published by OECD, SANTE, or EPPO are available for testing of substances used in plant protection. Many of these guidelines are not adapted for microorganisms. Risk assessment for biocides to enter the European market is described in the Uniform Principles, Commission regulation EU 546/2011 Part II. Risk assessment approaches for plant protection products are acceptance’s criteria that are not developed for microorganisms. Thus, no consistent approach is available in the different member states. This leads to uncertainties and non-acceptance of submitted data. We will discuss the current challenges in interpretation of the data requirements and propose solutions for the risk assessment of biocides based on microorganisms.

TUPC10
Ecotoxicological testing to support the assessment of Microbials
Biopesticides are an excellent alternative to chemical pesticides, and there is an increasing demand in testing and evaluation of these products. This poster focuses on microbial pesticides based on bacteria, fungi, viruses or protozoans as their active substances. Possible adverse effects to non-target organisms (NTO) are rather limited due to the narrow and specific host range of these microbial pest control agents. However, a complete risk assessment demands testing of NTOs, when exposure and risk cannot be fully neglected. The assessment of microbial biocontrol agents (mBCAs) and microbial biological control products (mBCPs) is relatively new and approved testing methods are not yet available in the same extent as they are for chemical pesticides. Not only the toxicity, but also the potential pathogenicity/infectivity needs to be addressed. Currently, the data requirements for mBCAs and mBCPs are Issue B of the European regulations 283/2013 (mBCA) and 284/2013 (mBCP). Numerous data requirements listed in these annexes were transformed directly from requirements for chemical pesticides and often cannot be adapted to the biological properties of mBCAs. In order to address the data requirements in a feasible manner, the biological properties of the microorganisms have to be taken into account, instead of strictly applying to current test guidelines. It is important to note, that testing is strongly influenced by physico-chemical properties of mBCAs. Microorganisms, i. e. with frequently used co-formulants, are not soluble which results in alteration of the test conditions (i.e. turbidity, O₂-demand, spray layers). Furthermore, organic components of the formulated product (i.e. yeast, starch) may lead to increased fungal growth in soil or test media. Additionally, the need to test at high concentration levels, lead to negative effects of partners (i.e. spores or co-formulants like kaolin) on the test organisms which are not related to the active substance and are difficult to interpret. Differences between OECD and OCSP (formerly OPPTS) guidelines, and requirements of the analytical verification in the test medium are addressed as part of the development of alternative ecotoxicological testing approaches. The findings of our ecotoxicological expertise presented in this poster can be considered as basis for further discussion in proposing different test designs addressing mBCA and mBCP requirements.

TUPC11
Microbiological Quantification Methods for MPCAs - Applicability to a Range of Microorganisms and Different Substances
M. Zetzmann, F. Kümmlich, A. Dabrunz, C. Lang, Eurofins Agroscience Ecots GmbH / Aquatic Ecotoxicology
In the last decade the number of biopesticide registrations in the EU and US have steadily increased. In the EU biopestices are regulated as plant protection products under regulation 1107/2009. Biopesticides cover a wide spectrum of substances including microbial pest control agents (MPCA) defined as products containing microorganisms (e.g. bacteria, fungi, protozoa, viruses). As for chemical plant protection products, regulatory authorities require an analytical verification of the doses applied in ecotoxicological tests also for MPCAs. Guidance can be derived from SANCO/3030/09 rev.4 and OPPTS 885.1400 (1996), but verification procedures need to be adapted on a case by case basis, as each microorganism possesses its own chemical properties and different growth conditions. Just as chemical methods, microbiological methods need to be validated for each specific microorganism. Experimental data will be presented with focus on the applicability of microbial quantification methods considering different microorganisms and substrates.

When ecotoxicology meets trophic ecology (PC)
TUPC17
Modelling bioaccumulation of persistent organic pollutants in Arctic food chains
R. Zondervan, Radboud University Nijmegen / Department of Environmental Science; A.M. Ragas, Radboud University / Department of Environmental Science; J.A. Hendriks, Radboud University Nijmegen / Department of Environmental Science
Persistent organic pollutants (POPs) are a group of chemicals with similar physical-chemical characteristics that are resistant to environmental degradation and biodegradation. Not only do these POPs bioaccumulate in the food chain, they are also known to cause adverse effects in fish, wildlife and humans. Although being banned in the previous century, many POPs are still present in high concentrations in Arctic areas, due to a combination of northward marine currents and their semi-volatile nature, high thermal stability and slow degradation turnover rates. As food webs in the Arctic are relatively simple, POP contamination may pose a great risk for animals at higher trophic levels, such as the polar bear (Ursus maritimas), hence the growing interest in studying bioaccumulation in the Arctic. Despite the large interest in bioaccumulation in Arctic food chains, the OMEGA model, as well as similar bioaccumulation models, are predominately validated on temperate food chains or relatively straight-forward Arctic food webs. In the present study, we aim to model bioaccumulation of multiple persistent compounds in the Arctic encompassing multiple species, using the OMEGA (Optimal Modelling for Ecotoxicological Application) bioaccumulation model. In this study, we aim to validate the model on Arctic areas by using a binning approach to include multiple species, in which species of a similar trophic level were binned.

TUPC18
Distribution and Trophic Magnification of Dechloranes, HBCDs, PCNs, and Other Legacy POPs in the Maritime Antarctic Ecosystem
J. Kim, Korea Polar Research Institute / Division of polar environment; Y. Choi, POSTECH Pohang University of Science and Technology; M. Barghi, POSTECH; J. Kim, J. Jung, Korea Polar Research Institute; Y. Chang, POSTECH Pohang University of Science and Technology
This study investigated distribution and trophic magnification of emerging persistent organic pollutants (POPs), including PCNs, HBCDs, Dechloranes, polychlorinated biphenyls (PCBs) and organochlorine pesticides (OCPs) in the maritime ecosystem in King George Island, Antarctica. The samples were collected in the Baton Peninsular in King George Island, Antarctica. from December 2013 to January 2014 and included Antarctic cod, icefish, limpet, amphipods, leopard seal, Gentoo penguin, Chinstrap penguin, kelp gull, and south polar skua. PCNs, HBCDs, Dechloranes, DDTs, HCHs, Pentachlorobenzene (PCBz), Hexachlorobenzene (HCBz), Chlorodanes, PCBs were detected in all samples, and the levels were the detection rates for the legacy POPs were more than 90 %, but those of some new POP compounds were only 50%. The detected POP levels in this
study were much lower than the levels in the previous study in low and mid latitude region, and even those in the Arctic. The trophic magnification factor (TMF) of each POP compound were calculated based on the ratio of stable isotope nitrogen and the log-transformed POP concentrations. Some of the compounds, OCs, HBCDs and highly chlorinated PCBs and PCNs, showed significantly positive correlations, suggesting biomagnification of the chemicals. DPs, however, showed insignificant results. After the TMF analysis for aquatic and terrestrial food web models, TMF values showed different trends compared to the TMFs in whole sample model. The inclusion of migrant animal, such as south pola skua and kelp gull, also arose an uncertainty to evaluate TMFs. The result of this study presented widespread contamination of the Arctic Environment by the New and Legacy POPs. The levels of most POPs were magnified through trophic levels, while Dechloranes, emerging contaminants, appeared not to enough TMF values. The insufficient detection rate of Dechloranes, complexity of the food web structure, and the overestimation due to migrant animals arose the uncertainties in TMFs, and therefore need to be taken into consideration to interpret the TMF results in this study.

TUPC19
Bioconcentration as the predominant mechanism for fish PCB contamination in alpine lakes.
T. Massel, Universite Savoie Mont Blanc; M. Perga, Universite of Lauzanne / Faculty of Geosciences and Environment; N. Cottin, Universite Savoie Mont Blanc; S. Cachera, CISALB; C. Piot, E. Naffrechoux, Universite Savoie Mont Blanc.

Bioconcentration and biomagnification relative contribution to the PCB burden in freshwater fish in alpine lakes ecosystems remain a debated issue. The aim of this study was to identify the relative role of those different processes for two fish species Coregonus lavaretus (European whitefish) and Salvelinus alpinus (arctic char) in one of the heaviest PCB contaminated alpine ecosystem: lake Bourget (France). The 7 indicator-PCB concentration and lipid content of fish filets were measured in European whitefish (n = 89) and arctic char (n = 55) from 2013 to 2016. Potential explanatory variables for differences in PCB contamination levels in fish were chosen to identify the impact of living and feeding habitat (using δ13C) and the influence of trophic parameters using δ15N and body size. Results showed a decrease of PCB burden in fish after the clean-up of the major input source of PCB in the lake and a steady situation since then. Arctic char was found to be significantly more contaminated than whitefish with a mean concentration of 25±132 ng.g
w/w and 45±28 ng.g
w/w respectively. Individual’s PCB contaminations in both species were not tied to feeding habitats (p>0.05). Trophic position (characterized with δ13C) was also not correlated with intra-species contamination variabilities for the char and was only slightly positively related to concentration variabilities for whitefish (p=0.04), dismissing the importance of biomagnification as PCB accumulation process. However, fish body size seemed to be a potential explanatory variable for individual’s PCB concentration variabilities for whitefish (p=0.002) and whitefish (p=0.10). This last observation could be explained by fish/water partitioning equilibrium to be reached, where fish would tend to accumulate more PCB through their lifetime, highlighting the effect of the bioconcentration process. A lower clearance rate due to changes in physiological parameters (lower gill/body weight ratio, lower metabolism and/or excretion rate) could also be involved.

TUPC20
The role of diet and age: organohalogen accumulation in an avian top predator
M.E. Losseth, The Norwegian University of Science and Technology / Biology; N. Brieis, Norwegian University of Science and Technology / Biology; I. Uelaurs, University of Antwerp / Biology; T. Nygård, T.V. Johnsen, J.O. Bustnes, Norwegian Institute for Nature Research NINA; D. Herzke, NILU - Norwegian Institute for Air Research / FRAM Centre Tromsø; G. Poma, G. Malavasan, University of Antwerp / Toxicological Center; A. Covaci, University of Antwerp, Toxicological Center / Toxicological Centre Dep of Pharmaceautical Sciences; B.M. Jenssen, Norwegian University of Science and Technology / Biology; V. Jaspers, Norwegian University of Science & Technology / Biology Occupying a high trophic level, the white-tailed eagle, Haliaeetus albicilla can accumulate a wide range of organohalogenated contaminants (OHCs), even at an early age. Their diet consists mainly of fish and seabirds; thus, a long food chain potentially resulting in biomagnification of OHCs. The nests can be exposed to high levels of OHCs through maternal transfer from the egg, and later through the diet. As nestlings develop and grow, concentrations of maternally derived contaminants are higher for the biological ages of the young and increase in body mass when monitoring OHCs in nestlings. The aim of the present study was to investigate how differences between years, locations and dietary tracers can explain variation in OHC accumulation in plasma of WTE nestlings. Stable isotopes (SI) of nitrogen (δ15N) and carbon (δ13C) were applied as proxies for trophic level and dietary carbon source, respectively. In addition, we included the possible confounding effects of age and body mass on the contaminant variation. Samples were obtained in 2015 and 2016 from 70 WTE nestlings from two archipelagos in Norway, Smøla and Steigen. In total, 14 polychlorinated biphenyls (PCBs), 7 organochlorinated pesticides (OCPs), 5 polybrominated diphenyl ethers (PBDEs) and 8 per- and polyfluoroalkyl substances (PFASs) were quantified in over 50 % of the analyzed plasma samples at each location and year. The WTE is a marine top predator; however due to the topography of the island Smøla WTEs may feed on a mixed terrestrial and marine diet. According to our preliminary analyses, WTEs in Steigen are feeding on a slightly higher trophic level than WTEs in Smøla WTEs, and may consequently accumulate more of the ingested heavy OHCs. Though, in our analyses the SI values were only important in explaining variation in POPs but not PFAS levels. We also observed that age at sampling is an important factor, as legacy POPs are decreasing while PFASs are increasing with age. However, there are differences between years at each location with higher OHC concentrations for Steigen in 2015 and Smøla in 2016, not explained by age or diet. Our analyses also demonstrate large variations within nests at both locations, suggesting that siblings may not always share prey. We hereby emphasize the importance of ecological and biological variables when investigating OHCs in an avian top predator.

TUPC21
Fate of PAH, phthalates and their metabolites in an urban river food web
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Trophic magnification factors have been extensively assessed for persistent organic pollutants, but remain poorly studied for metabolizable pollutants and their metabolites. Polycyclic aromatic hydrocarbons (PAHs) and phthalate plasticizers are continuously released in urban rivers and are rapidly metabolised and excreted by freshwater organisms, thus limiting their bioaccumulative potential. Abiotic and biotic samples, from primary producers to piscivorous fish, were collected in an urban river and analysed for PAHs, phthalates and their metabolites. Stable isotopes of nitrogen were used to determine trophic levels and to calculate trophic magnification factors (TMF) of each compound and its associated metabolite. Our results highlight a trophic dilution (TMF < 1) of all PAHs and phthalates, meaning that predators were less contaminated than their preys. When taking into account the associated metabolites, total body burden of PAHs still declined with increasing trophic levels, confirming the rapid transformation and excretion of these compounds within organisms, and a very limited trophic transfer. In contrast, the level of phthalate compound and its associated metabolite(s) increased from prey to predators, suggesting a lower clearance rate of phthalates and a slight biomagnification potential across freshwater food webs. At the light of these results, it appears essential to consider phthalate metabolites, instead of phthalate diesters only, in environmental risk assessment.

WPEC01
Does pre-exposure to bisphenol A affect the susceptibility of breeding zebrafish upon re-exposure?
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Bisphenol A (BPA) is a commercial important chemical used in the production of widely used epoxy resins and polycarbonate plastics and it is ubiquitous in the environment, resulting in widespread exposure of humans and wildlife. BPA was shown to cause reproductive effects via disruption of both the oestrogen and androgen signalling pathways. Recent studies suggest that BPA also affects epigenetic signallings pathways, including alterations in transcription of DNA methylation maintenance enzymes and altered DNA methylation profiles. This study aims to investigate how previous exposure of adult fish to BPA affects their response and the response of their offspring upon re-exposure, and whether there is an epigenetic basis for these effects. Breeding groups of zebrafish (Danio rerio) were exposed to 10 and 100μg BPA/L for 5 days, either once (C-10, C-100) or twice (10-10, 100-100) with a 13 day period of depuration in between, and appropriate controls were maintained in parallel. The adult gonads were sampled for transcriptional analysis. Reproduction was quantified over time, and embryos from each treatment group were then exposed to a range of BPA concentrations from 0-72hpf to measure their susceptibility to BPA exposure. There were no effects on reproductive output under our exposure conditions. However, at the transcriptional level, anti-Mullerian hormone (amb) was significantly downregulated only in fish that were re-exposed to BPA (100-100). In addition, embryos originating from adults which received a pre-exposure to BPA (100-100) were significantly more tolerant compared to embryos originating from
naive adults which received a single exposure to BPA (C-100). This suggests that pre-exposure of adult fish leads to a protective effect on their offspring. We hypothesise that these effects may be due to physiological changes or epigenetic memory between the first and second exposure period, and we will now change the promoter DNA methylation of amth to investigate this hypothesis.

WEPC02 Zebrafish as a model to assess transgenerational effects of environmental stress via epigenetic inheritance J. Kamstra, NMBU / BaSam Although toxicological effects of anthropogenic stressors present in the environment are studied extensively, few studies assess the epigenetic effects of such stressors and if these stressors can transfer these effects over multiple generations. It is hypothesized that some stressors are able to change the epigenetic state in germ cells causing effects on histones modifications, DNA methylation and miRNA expression, potentially inherited by subsequent generations. Here, the aim was to elucidate the role of epigenetics in transgenerational inheritance of effects of different stressors by measuring all epigenetic layers using the zebrafish model. Transgenerational studies were performed with three different stressors; a DNA methylation inhibitor, 5 azacytidine (5AC); a phthalate metabolite, mono2-ethylhexylphthalate (MEHP) and ionizing radiation. We employed state-of-the-art techniques to assess effects in multiple generations of zebrafish embryos and larvae at all epigenetic layers, but most extensively on DNA methylation. Following early life exposure to 5AC and MEHP, many changes of DNA methylation were found in the offspring, and these changes could be linked to gene pathways that are associated to those compounds, such as embryonic development and obesity. Subsequent analysis in two following generations lead to the conclusion that some regions were persistently changed. Concerning ionizing radiation, in F1 embryonic offspring from irradiated parents, many changes of DNA methylation were observed. These changes could be linked to effects that were found in the F0 generation such as DNA damage. Follow up analysis in the second and the third generation, revealed persistent effects of DNA methylation in a number of regions. Additionally, miRNA analysis in the offspring revealed a number of differentially expressed miRNA linked to similar pathways as with the DNA methylation dataset. Finally, changes in histone modifications were found at specific loci, but these changes were not observed in the second generation. Our results reveal considerable effects on DNA methylation following exposures during early life in zebrafish to MEHP, 5AC and ionizing radiation, as well as a role for miRNAs and histone post translational modifications for the latter. By linking the DNA methylation data to genes, results indicate a functional role for DNA methylation in zebrafish. Persistent effects in F2 and F3 implies that DNA methylation changes can be inherited by multiple generations.

WEPC03 Can changes in DNA methylation be linked to exposure of plants to radiation over multiple generations? N. Horemans, Belgian Nuclear Research Centre (SCK-CEN) / Biophase Impact Study, SCS-CN / E. Emeef, SCK-CEN / M. Vangeel, SCK-CEN / Biosphere Impact Studies; S. Gaschak, Chornobyl Center; K. Nanba, Institute of Environmental Radioactivity; R. Nauts, SCK-CEN In this study the impact on plants of long-term (transgenerational) exposure to radiation coming from nuclear accidents like Fukushima and Chernobyl is investigated and compared with lab experiments in either a chronic, acute, multi-, or single exposure. In a transgenerational set up, changes in DNA methylation could be the basis of transgenerational changes found in field or lab conditions. A field campaign was performed in both Chernobyl (CEZ) and Fukushima affected areas (FEZ) in the course of May 2016. Annual Brassicaeae plants, Arabidopsis thaliana and Capsella bursa pastoris in CEZ and FEZ, respectively, were sampled alongside a gradient of enhanced radiation ranging from 0.5 to 50 µGy.h⁻¹. Seeds from Arabidopsis thaliana were harvested in the CEZ and grown for one clean generation under lab conditions to score for multigenerational effects. In addition further lab experiments were performed on wild type plants of Arabidopsis thaliana grown under chronic exposure conditions (at 1 mGy/h) or more acutely (20 to 400 mGy/h) for 14 days in one, two or three generations. Plants were scored for their development, photosynthetic activity, and oxidative stress markers as well as germination rate and root growth. In general higher differences are found in plants exposed in a multigenerational setup than in a transgenerational one. The field plants did not show any abnormalities that could be correlated with the exposure gradient although some delay in flowering was observed in plants from medium and high radiation levels. The level of total DNA methylation could not be linked to the radiation gradient present in the field but rather to differences in developmental stage of the collected plants. In lab-exposed plants however global DNA methylation showed a significant increase which was both dose and generation dependent. Significant changes in transcription of methylation regulating genes were also measured in the different generations. Highest differences were present in the S1 generation but seemed to be reduced in the S2 generation. Overall the data hint towards a role of methylation in the response to radiation but its use as marker of exposure or in risk assessment needs further experimental evidence and discussion. This work was supported by European project COMET (7th PCRD EURATOM Contract Number: Fission-2012-3.4.1-604794) (www.comet-radioecology.org)/generation.

WEPC04 Evolutionary toxicity: tools to understand impacts of past, present and future environmental contamination S.F. Crawford, RWTH Aachen University / Institute for Environmental Research, Department of Environmental Toxicology and Evolutionary Genetics, RWTH Aachen University / Department of Ecosystem Analysis; M. Hinderer, Technische Universität Darmstadt / Institute for Applied Geoscience; A. Schwab, Technische Universität Braunschweig / Institute for Geosystems and Biosimulation; H. Holler, RWTH Aachen University / Institute for Environmental Research This research will utilize environmental reconstruction methods along with paleoecological, paleontological, and paleogenomic techniques to understand historical, current and potentially future trends in environmental contamination and associated impacts on lake systems. Long-term exposure to environmental contaminants can cause genetic adaptations in exposed populations of aquatic organisms. The new research fields of evolutionary toxicology and resurrection ecology offer powerful tools for the investigation of changes in sensitivities and adaptive trajectories of populations exposed to contaminants and environmental stressors over decades to centuries. Dormant resting eggs produced by Daphnia species (Crustacea: Cladocera) as a result of unfavourable environmental conditions are archived in sediments and can be dated and hatched to produce clonal lineages (i.e., same genotypes) of historical populations. This talk will present an overview of the evolutionary tools available and their current and potential applications in toxicological investigations. Additionally, we will present our preliminary research, which examines how genotypes of clonal lineages of Daphnia species from single populations, separated through generations of evolution, differ in their response to exposure of stressors. Results will provide insight into the sensitivity and fitness of organisms in response to environmental contaminants and the micro-evolutionary adaptations of genes that evolution in response to contaminants may result in. Furthermore, evolutionary changes in aquatic organisms can also result from other environmental stressors, such as temperature. Since increased temperatures are expected to occur in the future as a result of predicted climate scenarios, it is important to examine the fitness of historical and recent clones of Daphnia to temperature changes in combination with exposure of contaminants. Toxicological assessments and genomic data obtained from these archives of natural populations will provide unprecedented opportunities to gain insight into long-term and potentially future evolutionary responses of natural populations to environmental changes resulting from environmental stressors, including contamination and climate change.

WEPC05 Chemical and physical stressors shape the population genetic structure of aquatic invertebrate populations P. Inostroza, University of Gothenburg / Effect Directed Analysis; I. Véra-Escalona, Dalhousie University / Biology; A. Wicht, Eberhard Karls Universität Tübingen / Chemistry; M. Krauss, W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect Directed Analysis; H. Norf, Helmholtz Centre for Environmental Research GmbH - UFZ / River Ecology Aquatic Ecosystems Analysis and Management Organisms are rarely exposed to only one single stressor in the environment, but rather to multiple human-derived threats working simultaneously. Environmental pollution can modify population genetic structure via ecological bottlenecks, fragmented distribution of species to altered gene flow patterns, or increasing mutation rate. Organic micropollutants such as pesticides, biocides, pharmaceuticals, personal-care products, or industrial chemicals are ubiquitous in the aquatic environment and their effects are considered a relatively new and emerging anthropogenic pressure over evolutionary processes, especially potential effects of pollutants on genetic population structure may be more disruptive regarding ecosystem functioning than individual-level effects. Despite the bunch of investigations on genetic variation in wildlife, our understanding about the individual stressor effects on genetic variation is still limited. Recently, there has been an increased interest to integrate environmental chemistry and evolutionary toxicology approaches into the assessment of direct and indirect effects of anthropogenic stressors on genetic variation. To address these challenges the genetic structure of a shredder invertebrate, Gammarus pulex, was examined using evolutionary toxicology and body burden of organic micropollutant approaches. Exposure to chemical pollution alone and in combination with the presence of weirs resulted in a depression of allelic richness in native G. pulex populations. Our results suggest that the input of a mutagenic effluent from a WWTP resulted in a strong increase in private alleles and in genetic differentiation. In addition, the presence of weirs disrupted the migration across the river and thus the gene flow between G. pulex upstream and downstream. This study provides strong evidence that the assessment of genetic variation including private alleles together with the contamination of mutagenic and nonmutagenic chemical pollution offers new insights into the regulation of genetic population structure and highlights the relevance of emerging anthropogenic pressures at the genetic level.

WEPC06 Histone methylation as exposure biomarker of environmental chemicals
What's your take on communication? Don't Panic! Reports on how to accurately communicating science and risk (PC)

WEPC07  
Dangers misconceptions - Consumers need help!  
U. Klaschka, University of Applied Sciences  
Previous surveys revealed that average consumers and even more illiterate persons are struggling with risk communication instruments for harmful substances in commodities. The majority of consumers do not understand risk communication instruments as intended by legislators or do not use them at all. In contrast, the present survey focused on 'best-case' consumers who are interested in the topic, have a good education or a good self-reported knowledge in chemistry. These 'best-case' consumers use preferentially hazard pictograms in accordance with the Regulation on Classification, Labelling and Packaging (86%), reports in the media (80%) and information printed on the products (77%) to learn about harmful substances in consumer products. Surprisingly, smartphone applications (<10%) and information by authorities (14%) were not indicated as frequently used information sources. Most respondents considered information published by consumer and environmental organizations (75%) and the hazard pictograms (74%) trustworthy. Interestingly, the respondents considered legislators (94%), consumers (75%), manufacturers (71%) and environmental and consumer organizations (61%) as responsible for risk reduction. It is alarming that many of these 'best-case' participants assumed that food (up to 62%), products with an environmental label (36%), natural personal care products (36%), homeopathic products (36%), natural pharmaceuticals (26%) and products without hazard pictograms (11%) would not contain substances harmful for health or the environment. Nearly one out of ten respondents did not know that consumer products can contain harmful substances. These results show that motivation and knowledge in chemistry help, but are not sufficient. Consumers need support to understand risk communication instruments they need support to understand which products might contain harmful substances, they need support to determine the impact of harmful substances in commodities and they need support for suitable risk reduction behavior.

WEPC08  
The European Union Observatory for Nanomaterials (EUON): A new platform for communicating information on the safety of nanomaterials  
A. Sumrein, European Chemicals Agency (ECHA); J. Holmqvist, T. Aitasalo, European Chemicals Agency  
The REACH and CLP regulations are two key regulations addressing the manufacture and use of chemicals and the impact of these chemicals on human health and the environment. Although these regulations do not contain explicit requirements for nanomaterials, the regulations nevertheless address all chemical substances, including nanomaterials. In addition, over the past years, significant reformation on markets and safety aspects of nanomaterials in the EU market. In spite of this, there is a perception that there is insufficient information available in the public regarding the safety of nanomaterials. As a result, the European Commission entrusted ECHA with the creation, management, and maintenance of the European Union Observatory for Nanomaterials (EUON) [1] via a delegation agreement in December 2016[2]. The aim of the Observatory is “to give objective and reliable information on markets and safety aspects of nanomaterials in the EU market”. The presentatin will provide an overview of the activities of the EUON, including the background, the current content of the Observatory, and planned future developments.

WEPC09  
Roadmap for the unknown  
M. Luitwiler, M. H. Wegelmans, Biocearc earth  
The main environmental themes have been addressed in the last decades. Think acidification, eutrophication, nutrients and bulk industrial chemicals for which environmental guideline values have been derived within legal frameworks. Changes are ongoing in the scale level at which environmental problems are regarded as well as the scale level of industrial production. In the past large volumes of bulk chemicals were produced, now and in the future lower volumes of more specialised compounds are and will be produced. That means that more and more compounds will enter the environment in low volumes. Also time-to-market of new developments and technologies decrease which leaves less time for a thorough risk assessment. last but not least, techniques for measuring compounds are improving. More and more compounds can be measured in increasingly low concentration while the risks of these compounds in low concentrations are not known or just being studied. For the Province of Groningen en the Water Company Groningen the reason to develop a policy for emerging contaminants in the environment with the main question: What to do when emerging contaminants are found in soil or water. Biocearc earth has developed a roadmap for policy and/or actions by the province, municipalities, water company and water boards for emerging contaminants in the soil-water system. Our framework was: compounds and organisms that enter the environment or can be spread by human actions and that consequently have a negative effect on humans, nature or agriculture and for which no guideline values have been derived within the Soil Law, Water Law (Water framework directive) and Drinking water decree. The first step in the roadmap is to determine if a compound or organism in the environment can cause an actual or large risk. If no risks are present, no further investigation is needed. In all other cases, the roadmap needs to be followed. In the presentation we will devolve the process that has been followed to come to this roadmap as well as the background information. In the roadmap we describe the role of different stakeholders, including communication, enforcement, measurements to further prevent contamination or spread. In the presentation these roles will be further highlighted. Additionally we will organise a workshop regarding to discharge in January for province, municipalities, water boards, water company and RWS. The results of this workshop will also be highlighted.

WEPC10  
EVOKEO: enhancing the value of climate data - translating risk and uncertainty utilizing a Living Labs approach  
A. M. Deng, Norwegian Geotechnical Institute / Natural Hazards; L. Van Well, M. Zetterlund, Swedish Geotechnical Institute; G. Ellen, R. van der Brugge, DELTARES; J. Koerth, B. Vollstedt, Christian Albrechts University of Kiel  
The impacts of climate change are broad and although much focus has been on disaster risk reduction and coastal management, climate change will also have consequences for environmental management where the transport of contaminants, organism acclimation and vulnerable communities will be important to consider for future human and ecological risk assessments. In this context it is useful for environmental scientists to be familiar with the concept of climate services which are defined as the transformation of climate-related data into products, assessments, best practices) in relation to climate that may be of use for the society at large. Since the climate service sector is relatively new, there is a need to engage knowledge providers, users, and translators to identify improvements to climate services through co-design, co-development and co-evolution. The EVOKEO project aims to address this challenge by re-framing the risk and uncertainty associated with climate data into knowledge products more understandable and useful for end-users concerned with risk mitigation and adaptation. The project team will engage end-users in a Living Labs approach to ensure a user-contribution innovation methodology at established case study sites in Norway, Sweden, Germany and the Netherlands. The Living Lab methodology has emerged in recent years as a form of experimental and potentially inclusive mode of urban planning. Although the scope and character can vary depending on the issue at hand, the international level and the nature of the problem, the general idea is to involve a range of committed stakeholders in a real-life “laboratory” setting to test and develop alternative solutions for complex challenges, such as climate adaptation. The first activity for the Living Labs at each case study site will be a co-design process to encourage stakeholders to share their perceptions of risk and uncertainty. Since there are many different definitions and interpretations of risk, understanding these perceptions of risk is a prerequisite for communicating risk. Thus, EVOKEO supports the development of the field of climate services to improve our capacity to manage climate-related risks.

WEPC11  
Communicating monetary values of environmental impacts - case studies related to ISO DIS 14040  
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Monetary valuation of environmental impacts from human activities is a way of understanding and communicating its societal significance. However, monetary values are easy to accept without knowing the many ways they can be determined and the many perspectives they may represent. ISO TC 207/SC1 has set up a working group to develop a framework standard on monetary valuation of environmental impacts (as well as related aspects (sems) and use of resources) to increase transparency and its use in management. The standard contains requirements and recommendation on how to document and report information (metadata) about what a monetary value represents and how it is developed. As a part of the Swedish contribution to the work, three case studies were made to test the framework and to find out which metadata that had the greatest influence on decisions made. The cases represented three choices of ways to produce energy: (a) one between natural gas and waste heat from waste incineration, (b) one between different vehicle propulsion techniques, and (c) one between different types of sludge treatment and energy recovery. We have used the EPS 2015 dx method to value emissions and resources and a national Swedish database used for cost benefit studies. The results indicated that the framework has important merits. The report is about the system boundaries of the impact valuation which is the basis on which environmental goods and services that is included in the valuation. The system boundaries of impacts may vary in time, and object that is valued. The object may be chosen anywhere in a cause-effect chain. System boundaries also exist for the population whose values are assessed, and for the emissions and resources used. Other mega trends have public influence. The choice will discuss the other assumptions relating to future conditions. The cases, where the alternatives mainly differ due to more or less use of fossil fuel is rather insensitive to how the valuation was made with respect to the ranking of alternatives. In cases where there is a trade-off between use of fossil resources and scarce metals, the choice is very sensitive to the temporal system boundary of the impacts and affected population. A database format is proposed for documenting monetary values and related metadata.

WEPC12 Full STEAM Ahead: Merging Science and Communications to Investigate Environmental Questions G.K. Bildmeyer-Fraser, Jacksonville University / Chemistry; A. Kent-Willette, Jacksonville University / Communications; M. Simmons, Jacksonville University / Biology and marine sciences This project involved a case study and best practices surrounding successful STEAM interdisciplinary research. The collaboration was fostered through events and benefits studies. No results from a two year research agenda. The study focused on graduate and undergraduate interdisciplinary research in the fields of Environmental Science and Communications. Specifically, the influence of changing land use along the lower St. Johns River, Fl. was investigated, and the project and resulting data were published using modern communication tools such as social media, in tandem with more typical scientific means such as presentations at conferences and journal articles. The project and collaboration lead to applications and ultimately secured funding, successfully incorporated service learning and research opportunities for students, pursued and communicated meaningful research and managed teaching across very different disciplines.

WEPC13 Let's go visual, a picture is worth a thousand words: How to explain Emerging Contaminants using animations N. Ospina-Alvarez, S. Schneider, University of Potsdam / Institute of Earth and Environmental Sciences The development of new technologies has enhanced the use of several elements in information and communication technologies, semiconductors, electronic displays and 'green energy' related technologies. Platinum, indium, thallium are good examples of those kind of elements, that during long time were laboratory curiosities but that now have an important place as raw materials in high-tech products (optics, electronics, medicine). However, many of these elements are toxic (e.g. thallium, chlorine). Environmental processes can be used to test the hypothesis that increasing its availability in the environment. Basic research about Technology-Critical Elements (TCE) and Emerging Contaminants (EC) is needed, but also is part of the research process to transfer this knowledge to a general public. Within this framework, the University of Potsdam and GeoEd (http://geoeducation.de/) started a pilot project to develop teaching and learning material related to emerging contaminants. The goal is to use the project to show how a complex topic, can be easily included in modern science classes, going from a general concept (TCEs and EC) to a particular study case (phytoextraction of thallium from soils using mustard plants). All the material produced implements the Open Educational resources (OER) concept, which aims towards free access, documents with open license and media useful for teaching, learning, as well as for research purposes. The OER concept allows to new initiatives and projects, produce educational material accessible without time-wise or spatial barriers. Acknowledgement: This project is supported by an Outreach Grant of AXA Research Fund (Paris, France) and the Research Focus of Earth Sciences (RFES), University of Potsdam (Germany). Key-words: Emerging Contaminants, Technology-Critical Elements, raw materials, science animations, outreach.

WEPC14 Improving transparency, consistency and efficiency of ecotoxicological testing centres: developing text-based practice guidelines in ecotoxicology C.A. Geest, Vrije Universiteit Amsterdam / Ecological Science; N. van Straaten, Vrije Universiteit Amsterdam / Department of Ecological Science; T. Hamers, VU Amsterdam University, Institute for Environmental Studies (IVM) / Department of Environmental and Health; S. Moes, Vrije Universiteit Amsterdam / UBVU; M. Kraak, University of Amsterdam / IBED-FAME; J. Parsons, University of Amsterdam / IBED-ELD; S. Droge, University of Amsterdam/IBED Institute / IBED; J. Hermens, Utrecht University / Institute for Risk Assessment Sciences; M.G. Vijver, CML Leiden University / Conservation Biology; N. van den Brink, Wageningen University / Dept of Toxicology; A.M. Ragas, Radboud University / Department of Environmental Science; A. Lühr, F. van Belleghem, Open University / University of Technology.

Although several textbooks are available, teaching environmental toxicology in general seems to suffer from a lack of a well-elaborated, up-to-date and consistent textbook that covers all aspects of the field. As a consequence, every university is developing its own training materials in addition to a textbook, but only little of this material is available online. And if materials are online, they are not consistent, lack novelty or do not cover the entire field. A Dutch consortium therefore took the initiative to develop an open online textbook on Environmental Toxicology that should cover the field in its full width, including aspects of environmental chemistry, ecotoxicology, toxicology and risk assessment. The initiative is sponsored by the Netherlands Ministry of Education. The project aims at developing an open online open book on Environmental Toxicology that is useful for training at BSc, MSc and higher levels. The book will be designed in a modular way, with each module having a clear training goal/attainment level and finished with a number of keywords. The book will also contain tools for self-study and training, like exercises and questions. With the book, we aim at improving quality, continuity and transparency of the education in environmental toxicology. We also want to make sure that fundamental insights on fate and effects of chemicals gained in the past are combined with recent approaches of effect assessment and molecular analysis of mechanisms causing toxicity. To guarantee quality of the book and associated training materials, we aim at having 1-2 authors for each module and also 1-2 reviewers from outside the team of authors. In addition, an advisory board will be involved in supervising the project, as well as educational advisors, while the project team will serve as an editorial board. The project team, consisting of a diverse group of (international) experts from 12 Dutch universities, does not possess all expertise to cover the width of the field. We therefore solicit contributions from as many colleagues as possible from within the SETAC community. With that, we hope we can produce a book that is written and supported by SETAC, that is covering the entire field, and is useful for training within e.g. the SETAC Europe Certified Risk Assessor (CRA) programme. The publication as an open online book will allow continuous updating of the book, providing a possible role of SETAC in sustaining the book.

WEPC15 Policy learning through professional forums in the field of environmental toxicology: What role for the Society of Environmental Toxicology and Chemicals (SETAC)? M. Mondou, McGill University - Macdonald Campus / Dept Natural Resource Sciences; G. Hickey, McGill University - Macdonald Campus / Natural Resource Sciences; S. Maguire, McGill University; D. Crump, Environment and Climate Change Canada / National Wildlife Research Centre; M. Hecker, University of Saskatchewan / School of the Environment & Sustainability and Toxicology Centre; N. Basu, McGill University / Faculty of Agricultural and Environmental Sciences Leveraging more than a hundred years of experience and knowledge (Gallo 2008), modern toxicology has crystalized in a set of highly codified and standardized practices that are used by industry and regulatory agencies internationally to assess the risk of chemicals to the environment. Toxicity testing methods using whole animal studies have long provided the general framework of instrumental beliefs concerning the most appropriate way to pursue the goals of environmental toxicology. While such in vivo methods are useful for determining the acceptable levels of single chemicals in the environment, they have a number of limitations that are broadly accepted within the profession. Most significantly, conventional in vivo methods are time consuming, can only be performed with a limited number of test animals and can take a considerable time and effort to produce meaningful results. As such, new alternatives have emerged to reduce the number of test animals required to detect toxic effects. Some of these include in silico methods. In silico methods are computationally intensive and require specialized knowledge to develop and implement. In silico methods are designed to be used in concert with traditional in vivo methods to reduce the need for animal testing and improve the efficiency of chemical testing. In silico methods can potentially reduce the number of test animals required to detect toxic effects, thereby reducing the number of animals used in chemical testing. However, the effectiveness of in silico methods in predicting the safety of chemicals in the environment remains uncertain due to the limitations of current models and databases. This uncertainty has led to calls for the development of more accurate and reliable in silico methods to support regulatory decision-making. In order to achieve this, it is essential to continue to develop and improve in silico models and databases and to validate their performance against experimental data.
Thinking green and circularly about microparticles, nanomaterials and composite materials: approaches for recovery, recycling and reuse (PC)

WEPC17
Biochar-mortar composites for construction materials
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Feasibility of biochar for construction material was examined through synthesis of biochar-mortar composites and evaluation of their construction and environmental properties according to mixing ratios. Characterization of biochar-mortar composites showed that 3-5% biochar inclusion did not significantly change their flowability, compressive strength, and thermal conductivity. As biochar content increased in biochar-mortar composites, benzene concentration in air was accordingly reduced, suggesting that biochar may be favorable to remove toxic contaminants causing sick building syndrome. Toxicity characteristics leaching procedure (TCLP) and Micotox® bioassay tests showed that biochar-mortar composite were not toxic. Our results suggest that biochar-mortar composites may be promising environmental-friendly materials for building and infrastructure construction area.

WEPC18
Complex Formation Trends of Ligand Binding toward In(III) and Ge(IV)
D. Wondrousch, G. Schuurmann, Helmholz centre for environmental research - UFZ / Department of Ecological Chemistry
In recent years, the demand for strategic elements such as Indium and Germanium has increased due to strong global economic growth, especially in the realm of semiconductors. Dwindling resources and growing demand necessitate new recycling strategies and the reassessment of existing repositories in the light of newly developed technologies. In this context, QSAR methods can be utilized in the development of chelating agents designed to remove toxic heavy elements. Through this, novel ligand concepts can be rapidly assessed and synthesis can be prioritized towards promising ligands, resulting in a shortened development cycle and reduced research costs. In our quantum chemical study, we analyze a systematic set of chelators with respect to their complex formation energies toward selected In<sup>3+</sup> and Ge<sup>4+</sup> complexes. Following a first principles approach, both Density Functional Theory and higher levels of theory have been used for the calculations, also addressing bulk solvation effects. The study focuses on both affinity and selectivity. General trends in binding affinity to selected ions are discussed as related to the electronic structure of the compounds. Chelator selectivity toward In<sup>3+</sup> and Ge<sup>4+</sup> is investigated in comparison to Fe<sup>3+</sup>, Fe<sup>2+</sup>, Cu<sup>2+</sup> and Zn<sup>2+</sup>. The importance of both properties arises from expected high concentrations of these interfering ions relative to the strategic elements of interest. Financial support from the Krüger Research School “Biodynamical and Geographical Elements” BHZM (Nr. 02110205) is gratefully acknowledged.

WEPC19
Cellulose Nanofibers as building blocks for innovative materials for remediation
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From the point of view of circular economy cellulose is one of the most interesting biopolymer since it derives from renewable sources which, thanks to its peculiar structural properties, it is widely used in the design of simple and advanced materials for different applications. Recently, cellulose nanofibers (CNF) were proven to be versatile building blocks for many preparations. The regioselective oxidation of the C6 primary hydroxyls of the anhydroglucose units, catalysed by TEMPO in the presence of the NaBr/NaClO oxidant system, leads to the formation of TEMPO-oxidized CNF (TOCNF), conferring to the cellulose many interesting properties. We recently report the synthesis of sponge-like nano-structured materials by cross-linking TOCNF and branched polyethyleneimine (bPEI). These materials were fully characterized from a chemical, structural and mechanical point of view. Quantitative information on their inner microstructure were collected by Micro-Computed Tomography (μ-CT) analysis. In addition the material can be easily modified in order to introduce additional chemical or structural properties. As an example, by functionalizing the bPEI with pNO<sub>2</sub>-phenyl-urea units it is possible to obtain a material that can be employed for the heterogeneous and selective aqueous release of fluoroanions. Furthermore, the addition of citric acid (CA) as co-crosslinker enforce the mechanical and structural performances. Another application of bPEI-TOCNF sponges is the controlled release of active principles. They have been used it for adsorption and release of two model drugs, amoxicillin and ibuprofen. The material showed very good performances in adsorbing both model drugs (~200 mg/g) from methanol solution. Interestingly, the presence of CA led to slower kinetic release in aqueous environments if compared with materials obtained without CA. The ongoing NanoBioN project is focused on the use of these sponge-like materials for soil and water remediation. We have demonstrated that these materials exhibit superior performances in removing contaminants both from fresh and from sea water. In particular we are able to remove heavy metals (Zn<sup>2+</sup>, Cd<sup>2+</sup>, Pb<sup>2+</sup>, Cr<sup>3+</sup> and Cu<sup>2+</sup>) and organic contaminants (e.g. p-nitroanisole). The knowledge regarding the effects obtained on contaminated organisms (Avelelas et al. 2017; Martins et al. 2017). Martins et al. (2017) showed no acute effects on marine clams (till 100 mg/L), but significant biochemical effects, even at low exposure concentrations. The present study aimed to assess the hazard of Zn-Al LDH in several marine species representing different trophic levels. Exposure tests were carried out with 15 species, including bacterium (Vibrio fischeri), cyanobacterium (Arthrospira maxima), microalgae (Isochrysis galbana, Nanochloropsis gaditana, Phaeodactylum tricornutum, Tetraselmis chuii, Mytilus galloprovincialis), polychaete (Heliste diversicolor), crustaceans (Acartia tonsa, Artemia salina and Palæmon varians) and sea urchin embryos (Paracentrotus lividus). Acute and short-term chronic exposure tests followed standard or well described guidelines, with appropriate adaptations in some cases. Exposure concentrations ranged between 0.01 and 100 μg/L depending on the species tested. The nanomaterial exhibited no toxic effects in most of the tested species, even at the highest exposure concentration. However, adverse sub-lethal effects, such as changes in the enzymatic activity or the physiological endpoints were observed in bivalves and crustaceans. We further provided the predicted no-effect concentration (PNEC) of Zn-Al LDH for seawater was set at 0.2 μg/L based on the lowest NOEC available (Martins et al. 2017). The results suggest that Zn-Al LDH is a promising engineered nanomaterial featuring a very low environmental hazard.

WEPC20
Zn-Al layered double hydroxides: a promising eco-friendly engineered nanomaterial
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Layered double hydroxides (LDH), also known as anionic nanoclay, are a class of inorganic engineered nanomaterials with a plate-like structure featuring a lateral size of 20-40 nm. LDH are characterized by positively charged metal hydroxides (e.g. Zn<sup>2+</sup>, Al<sup>3+</sup>) stabilized by anions (e.g. NO<sub>3</sub>) and water molecules between layers. LDH have remained a research area in aqueous environments if compared with nanomaterials obtained without CA. The ongoing NanoBioN project is focused on the use of these sponge-like materials for soil and water remediation. We have demonstrated that these materials exhibit superior performances in removing contaminants both from fresh and from sea water. In particular we are able to remove heavy metals (Zn<sup>2+</sup>, Cd<sup>2+</sup>, Pb<sup>2+</sup>, Cr<sup>3+</sup> and Cu<sup>2+</sup>) and organic contaminants (e.g. p-nitroanisole). The knowledge regarding the effects obtained on contaminated organisms (Avelelas et al. 2017; Martins et al. 2017). Martins et al. (2017) showed no acute effects on marine clams (till 100 mg/L), but significant biochemical effects, even at low exposure concentrations. The present study aimed to assess the hazard of Zn-Al LDH in several marine species representing different trophic levels. Exposure tests were carried out with 15 species, including bacterium (Vibrio fischeri), cyanobacterium (Arthrospira maxima), microalgae (Isochrysis galbana, Nanochloropsis gaditana, Phaeodactylum tricornutum, Tetraselmis chuii, Mytilus galloprovincialis), polychaete (Heliste diversicolor), crustaceans (Acartia tonsa, Artemia salina and Palæmon varians) and sea urchin embryos (Paracentrotus lividus). Acute and short-term chronic exposure tests followed standard or well described guidelines, with appropriate adaptations in some cases. Exposure concentrations ranged between 0.01 and 100 μg/L depending on the species tested. The nanomaterial exhibited no toxic effects in most of the tested species, even at the highest exposure concentration. However, adverse sub-lethal effects, such as changes in the enzymatic activity or the physiological endpoints were observed in bivalves and crustaceans. We further provided the predicted no-effect concentration (PNEC) of Zn-Al LDH for seawater was set at 0.2 μg/L based on the lowest NOEC available (Martins et al. 2017). The results suggest that Zn-Al LDH is a promising engineered nanomaterial featuring a very low environmental hazard.
volume of the microfibres released. In the current study, we assess the release of microfibres from different fleece fabrics with the aim of identifying production methods and fabric properties that release the fewest microfibres during domestic washing machines. A filter unit consisting of a 300 µm filter and a 100 µm filter in series was connected to the effluent pipe of a domestic washing machine. Each fleece test fabric (140cm x 90cm) was prepared by overlocking the edges to prevent lint from entering the filter and testing the total effluent. The fabrics were washed in a standard synthetic clothing program (28°C, 1 hr, 40°C). Weights inside the washing machine assured same mass for each material assessed and a consistent water flow into the machine. Effluent water was collected in a clean container and a sub-sample (1 L) passed through a 20 µm filter to collect any microfibres that pass through the filter unit. Each test fabric was first washed to study release in new clothes, and then washed a second time to determine release of the washed, and re-washed fabric. To improve the accuracy of the results, the two washing procedures were repeated in triplicate. A rinse cycle was run between each test wash to ensure removal of any remaining microfibres from the system. The hoses connecting the filters contained residual microfibres after washing; these were collected after each wash by cleaning the hoses manually. The pre-weighed filters were allowed to dry before the mass of fibres were determined. The number of fibres was then estimated based on microscopy counting of a pre-weighed sub-sample of the microfibres. Preliminary results show that ~80–90% of fibres in the effluent water are trapped by the 300 µm filter. Furthermore, the fibre release declines with successive washings. A detailed study of the underlying mechanisms is currently ongoing and the results will be used as a basis for ranking.


Nano-agochemicals promise higher efficiency than conventional pesticides, but much has to be learned about the gain of efficiency compared to conventional products, and the risk of directly applying such new types of yield enhancers on agricultural soil. Due to the relatively low acute toxicity and high natural abundance of silica nanoparticles (SiO$_2$-NPs), they are highly attractive for benign-by-design strategies in agriculture. Here, we present initial results of experiments that are in the process of being conducted on a laboratory scale to compare the effects of SiO$_2$-NPs and conventional fertilizer and pesticide ingredients, and combinations thereof, on the agricultural legume alfalfa (lucerene, Medicago sativa). The SiO$_2$-NPs used for the experiments were ~60 nm in primary particle diameter. As reference substances for conventional pesti- cides, the broadbanded fungicide tebuconazole was tested. Seed germination and infection tests, and a plant growth test were conducted. The Si was quantified by inductively coupled plasma – optical emission spectroscopy (ICP-OES). Beneficial effects of SiO$_2$-NPs were found for the fungal infection and germination rates in alfalfa, while the growth rates in the seedlings transferred to and grown in soil remained largely unaffected. The results confirm the moderate protective effects of silica nanoparticles on plants that have been reported previously, likely linked to the release of orthosilicic acid (Si(OH)$_3$) acting as a phytostimulative micronutrient. The use of silica in nanoagochemicals promises to reduce the organic pesticide burden of agricultural soil and crops. Acknowledgement - The authors thank the Swiss National Science Foundation (http://psf.nsf.ch/Project-168187) and the Adolphe Merkle Foundation for the support and funding of the study. We thank Laura Rodriguez-Lorenzo, Dimitri Vanhecke, and Sandor Balog for helpful discussions regarding ICP-OES analytics, electron microscopy, and dynamic light scattering, respectively.

WEPC24 LCA and beyond - integrating sustainability and/or other dimensions to improve decision support (PC)

WEPC25 Life Cycle Assessment of applying Algal Oil in salmon aquaculture: challenges for methodology and tool development H. Bosch, DSM Nutritional Products; A. Wojciechowski, Evonik Technology & Infrastructure; M. Binder, Evonik Nutrition & Care GmbH; F. Ziegler, RISE Research Institutes of Sweden

Evonik DSM founded the joint venture Veramaris®, introducing a new Algal Oil based omega-3 fatty acid source for aquaculture. This intracellular oil is produced in a biotechnological manufacturing process using non-marine resources. The rationale for this development is that the capacity to generate omega-3 fatty acids through fish is not sufficient to fulfill the dietary requirements of a growing population, and that many fish species used as feed in aquaculture are either fully utilized or oversupplied, leaving little room for expansion. Algal Oil reduces the dependency of salmon aquaculture on marine fatty acid production and fish stocks, by replacing marine ingredients with algal oil and crop-based ingredients. To illustrate the environmental impacts and potential tradeoffs of this new product, a Life Cycle Assessment (LCA) was performed. Indicators developed for application of LCA to fisheries were used in an LCA to assess the marine ecosystem impact of replacing fish meal and fish oil by Algal Oil in salmon feed. The analysis had to be performed in a separate calculation outside the LCA software, because the software does not include the required data and methods. The study demonstrated that the use of Algal Oil as a source of omega-3 fatty acids leads to a considerable reduction of impact on marine ecosystems of farmed salmon. This improvement is accompanied by an increase in impacts associated with agriculture. Current LCA methodologies do not allow weighting of these opposing effects. However, to make informed choices between the available options this would be required. Because the availability of natural marine resources is limited, the strong growth expected in salmon aquaculture requires innovative feed solutions decoupled from limited fish stocks to meet future requirements for omega-3 fatty acids. Veramaris® Algal Oil in combination with vegetable crops enables growth of salmon aquaculture that is independent of limited fish stocks. To support the choices discussed in this presentation, and similar choices, implementation of fishery impact assessment methods in LCA tools and development of weighting methodology is essential. Just as for other biotic impacts, there are also challenges of non-linearity and temporal and spatial variability connected to fishery-specific impacts, which are of a more local nature than global-scale life cycle impacts.

WEPC26 Balancing Environmental and Health Impacts of Food Production and Consumption C. Walker, Institute of Environmental Engineering, ETH Zurich; S. Hellweg, ETH Zurich / Institute of Environmental Engineering

An individual’s food choices can affect not only the magnitude of their food related environmental footprint, but can also have a direct effect on their personal health. People consuming lower amounts of meat tend to have lower environmental impacts, but it has also been found that they also tend to have lower risk of certain diseases. For the other hand, people with low vegetable or fruit consumption may also have relatively lower environmental impacts, while having increased risk of disease. This study investigates the daily eating patterns of a European population sample to identify and compare each individual’s environmental impacts due to their food production as well as the health impacts that can be expected due to their food consumption patterns. The Global Burden of Disease has identified dietary risk factors that have been proven to contribute to increased disease risk such as low fruit, vegetable, nut and seed, omega-3 intake and high red meat or processed meat intake. The relationship between the environmental impacts from producing foods classified in the dietary risk factor categories are compared to the health impacts associated with consuming these foods. From this investigation, we can estimate the magnitude of the health benefits associated with additional food production, as is shown in a case study of whole grain consumption. Results show that for individuals under-consuming whole grains (less than 125 grams daily), for every 1 micro disability adjusted life year (µDALY) increase in production impacts, there is a health benefit of 141 µDALYs. Similar results were found for all...
dietary risk categories in which under-consumption of a particular food group was considered a dietary risk. In cases where overconsumption poses a health risk, as is the case in certain meats, sodium, and sugar sweetened beverages, a 1 µDALY increase in production impacts is associated with increases in health impacts to varying degrees, ranging from 1.2 µDALYs for red meat up to 36.8 µDALYs for sugar sweetened beverages. This study found that for most of the dietary risk categories, health impacts due to consumption far outweighed the environmental impacts (measured in terms of µDALYs), however this study did not include environmental impacts as they are related to other impact categories such as ecosystem damage and resource depletion, which must also be considered to fully capture food production impacts.

**WEPC27**

What not to waste? Improving decision support for Food Loss and Waste (FLW) mitigation by considering food security and environmental sustainability

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LCA-based methods provide evidence of the largescale environmental impacts of food production. Nearly a third of food produced is lost or wasted, meaning production impacts occur with little to no societal service. The primary focus of response e.g. the UN SDG 12.3 has been on avoiding food loss and waste (FLW) quantities, for example halving food waste by 2030. Decision-makers, thus may prioritize FLW mitigation based on quantity, economical gain, and ease of implementation. To support multi-criteria sustainability decision-making we develop a framework and perform a global screening to prioritise FLW mitigation efforts based on two material issues: environmental impacts and nutrition security. LCA-based methods quantify environmental impacts related to FLW streams, and nutrition and global burden of disease data are used to quantify nutrition security. A global screening of FAO data on food production, supply, and FLW for various food categories (e.g. grains) is performed in 15 countries. Results demonstrate vastly different environmental impacts and nutrition security potential associated with various FLW streams. The results suggest that there is sufficient production of most nutrients globally, suggesting that in most cases food systems do not need to grow, but need to be optimized to reduce FLW and offer appropriate regional supply.

**WEPC28**

ARIADNA Project. Analysing the sustainability of implementing a mandatory Deposit-Refund System in Spain

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There is currently an ongoing debate in some Spanish regions on whether the implementation of a mandatory deposit-refund system (DRS hereafter) would be appropriate. A DRS can be simply defined as a system in which consumers pay a certain amount in concept of “packaging deposit” added to the price of a product and receive the refund back when they return the used packaging. This is a system in place in some European Countries such as Germany, Finland or Denmark whereas others such as France or the UK have refused its implementation. From a regulatory point of view, the Spanish Law 22/2011 (which transposes the correspondent European Directive 2008/98/CE), clearly states that the implementation of a DRS should be based on the analysis of its technical and economic feasibility, a set of environmental, social and health impacts and must ensure the proper functioning of the internal market. The study presented here follows this regulatory framework, analysing the sustainability of implementing this new system, combining environmental, economic and social studies in order to get an holistic picture of its feasibility. In particular, the following methodologies were used: (1) Life Cycle Assessment (LCA) for the environmental study; (2) Classic Cost Accounting for the economic study and (3) Social Footprint and Quantification of the Integrated Social Value for the social study. All of them applied using the same data, system boundaries and modelling restrictions. The study clearly concludes that the introduction of a DRS in Spain, under the studied conditions, is not advisable. This is because: a) according to several environmental indicators, although it could achieve an improvement in the overall recycling rate, it carries a higher environmental impact, mainly due to the duplication of collecting systems and the increase demand in transportation; b) represents a 4.6-fold increase in the economic cost to society (11 times if we only consider the management of DRS containers); and c) from a stakeholders’ point of view, it will have a significant increase both in space and time needs, and it will cause a higher Social Footprint.
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Development.

Ecological risk assessment.

Cytotoxicity.

Decision analysis.

Degradation.
Elimination.

Endocrine disruption.

In situ.

In situ.

In situ.

In situ.

In situ.

In situ.

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In situ.

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In situ.

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